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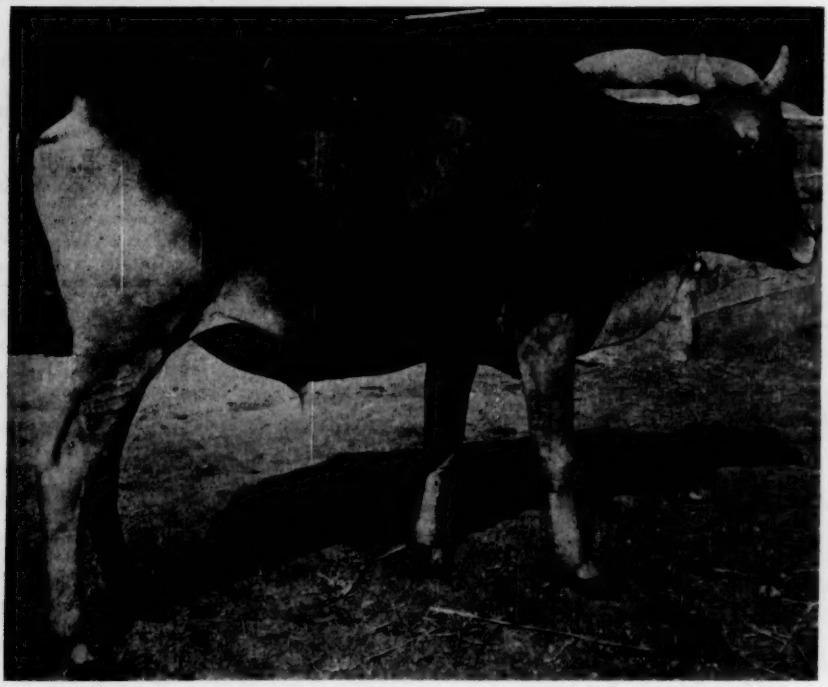

SHORT-TAILED CATTLE OF THAILAND

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Inbreeding in Andhra Pradesh

Complexities of Taste Variation

Dwarfism and Insulin Sensitivity in Cattle

The Relation Between Sex and Survival in Swine

VOLUME 51 NOVEMBER-DECEMBER, 1960 NUMBER 6

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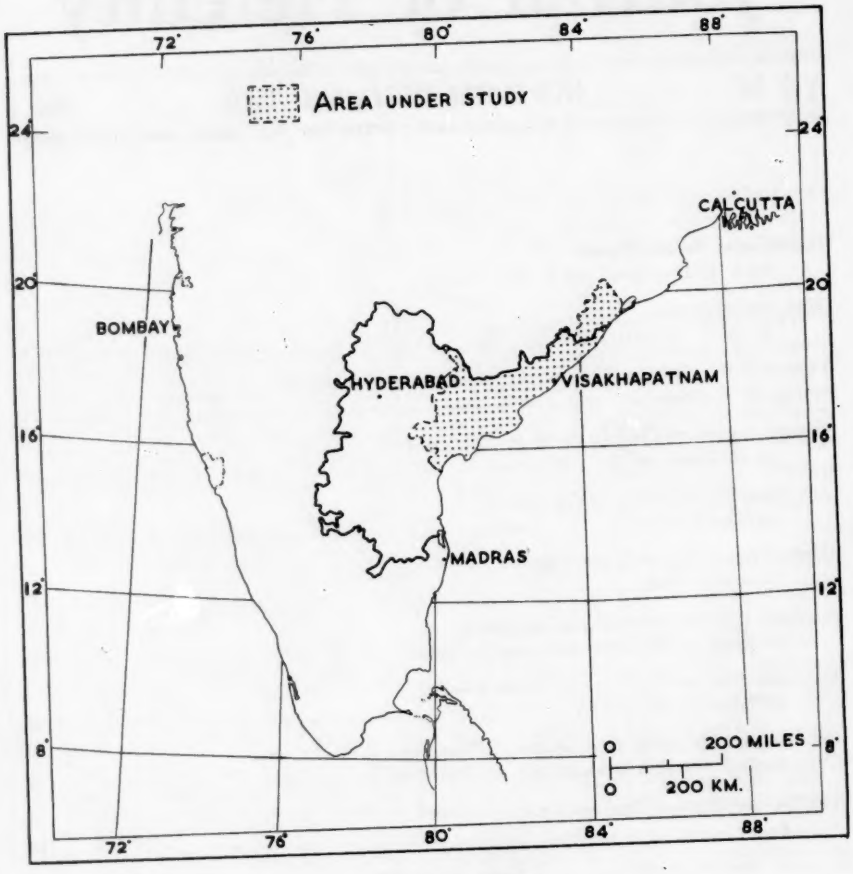

ANDHRA PRADESH

Figure 1

On this map of southern India, the dotted area is under study. It extends into the Ganjam district of the northeastern state of Orissa.

INBREEDING IN ANDHRA PRADESH

K. R. Dronamraju and P. Meera Khan*

MANY papers have been published on the rates of inbreeding in different countries of the world. Some of these, such as Ishikuni *et al.*[2] and Spuhler and Kluckhohn[5] dealt with the populations of some small islands where people tended to marry within the island community. These communities were described as *genetically isolated* populations and, as anticipated, showed a very high rate of inbreeding. According to Ishikuni *et al.*[2], in Hosojima, "the consanguinity rate appears to be the highest yet recorded for any group." In this paper, we record the preliminary results of a survey showing the highest consanguinity rate of marriages ever reported in a *mainland* population.

As the bulk of these data concerns caste Hindus, it should be emphasized that in this region each caste consists of such a large population that inbreeding does not occur because of lack of choice of marital partners. For example, in Andhra Pradesh there are more than a million people who belong to the Brahman caste. Hence, these caste groups are *much larger* "genetic islands" than those discussed by the authors cited above.

Material

There are about 33 million people speaking Telugu in southeastern India. The state of Andhra Pradesh was formed in 1956 on a linguistic basis to include the areas which they inhabit. The state is *not isolated* geographically from the neighboring states of India. Among Hindus the caste system is universal in the state, and most people marry within their own caste.

It has long been known both inside and outside India that Telugu-speaking people as well as other south Indians marry their close relatives[3]. We have begun a survey of the inpatients of King George Hospital in Visakhapatnam to obtain information about the frequency, nature and genetic effects of this inbreeding. The King George Hospital is a general hospital and is the largest in Andhra Pradesh, drawing patients from the thickly populated coastal districts of the state and the southern parts of the neighboring state of Orissa (Figure 1). Each of the patients was asked to give the degree of relationship between his parents, his own relationship to his spouse, and his married children's relationship to their spouses.

In this, the first sample obtained, all of the patients were female and were in the maternity and gynecological wards of the hospital. We are aware that the sample is biased. Out of 143 patients questioned two Hindus and one Muslim were unmarried. So we have information about 143 marriages of patients' parents, 140 marriages of patients, and 44 marriages of patients' children. A high percentage of consanguinity was found, as expected. Many of the Hindu patients and one Muslim who had not married a close relative explained, some of them apologetically, that they had been unable to do so because their elder

*Indian Statistical Institute, Calcutta-35, W. Bengal, and King George Hospital, Visakhapatnam-2, Andhra Pradesh, India, respectively. Mr. Dronamraju's present address: Department of Genetics, The University, Glasgow W. 2, Scotland. We are grateful to Dr. V. S. Raghunadhan for permitting us to conduct the investigation; and to Professor J. B. S. Haldane and Dr. H. Spurway for their many helpful suggestions. We also thank the Regional Survey Unit of the Indian Statistical Institute for help in providing the map of southern India.

sisters had married all the close relatives available, clearly stating that they would have preferred such a marriage. The frequency of inbreeding is high in Andhra Pradesh because the community regards certain relatives as socially desirable marital partners. Society equally forbids or disapproves of marriages between other relatives. The permissions and prohibitions differ between Hindus, Christians and Muslims.

Results

The consanguineous marriages fall into six groups on the basis of the type of relationship between the partners. The numbers of these marriages are given in Table I for Hindus, Christians and Muslims. The Hindus are further divided broadly into the four *varnas* or original subdivisions used in the *Vedas* (oldest literature in Sanskrit). Although people no longer follow strictly the professions which formed the basis of this classification, all but two of the patients mentioned the name of their caste or sub-caste) which is traditionally regarded as a subdivision of one of the *varnas*). These four divisions, which in contemporary Andhra Pradesh still show some differences in their ways of life, are the *Brahmans* (clerics), *Kshatriyas* (fighting men dependent on landed property), *Komatis* (the local name for the merchant caste called *Vaisyas* in Sanskrit) and *Sudras* (craftsmen including agriculturists). The *Harijans* (formerly untouchables) form a "fifth people" subse-

TABLE I. Frequencies of different kinds of consanguineous marriages

	Maternal uncle	Paternal uncle	Maternal uncle's son	Paternal aunt's son	First cousin once removed	Second cousin	Consanguineous total	Non-consanguineous	Consanguinity %
Hindus									
Sudras									
Parents	8	0	0	12	0	0	20	60	
Patients	7	0	3	10	5	5	30	50	34.06
Children	5	0	0	5	3	1	12	10	
Brahmans									
Parents	0	0	0	2	1	0	3	10	
Patients	0	0	2	3	0	0	5	8	22.50
Children	0	0	0	0	1	0	1	13	
Komatis									
Parents	2	0	0	2	0	0	4	11	
Patients	2	0	0	5	0	0	7	7	40.54
Children	0	0	0	3	0	1	4	4	
Kshatriyas									
Parents	0	0	1	2	0	0	3	7	
Patients	1	0	0	0	0	0	1	9	20.00
Children	0	0	0	0	0	0	0	0	
Harijans									
Parents	1	1	0	1	0	0	3	4	
Patients	1	0	0	1	0	1	3	3	46.15
Children	0	0	0	0	0	0	0	0	
No caste									
Parents	0	0	0	0	0	0	0	2	
Patients	0	0	0	0	0	0	0	2	0
Children	0	0	0	0	0	0	0	0	
Christians									
Parents	0	0	0	1	0	0	1	12	
Patients	1	0	0	2	0	1	4	9	19.23
Children	0	0	0	0	0	0	0	0	
Muslims									
Parents	0	0	0	0	0	0	0	3	
Patients	0	0	0	1	0	0	1	1	20.00
Children	0	0	0	0	0	0	0	0	
Totals	28	1	6	48	10	9	102	225	
Percentages	8.56	0.306	1.83	14.68	3.06	2.75	31.19	68.81	

quently accepted as Hindus by the members of the original four *varnas*. We explained these castes in the traditional order but in Table I they are arranged in the order of the number of marriages. Two patients who had made intercaste marriages stated that they had no caste.

The marriages of *Sudras* are about 61.5 percent of the total Hindu marriages. The rest of the caste groups are represented in much smaller proportions. There are 26 Christian and five Muslim marriages. The consanguinity rates among the *Harijans* and the *Komatis* are slightly higher than the others, but the differences between the castes among the Hindus, and between Hindus, Christians and Muslims are not significant.

The data about parents' consanguinity is interesting. The amount of inbreeding in the parental population is not significantly different from that of patients and children. But it is noticeable that no consanguineous marriage was recorded with a second cousin and only one each with a first cousin once removed and a maternal uncle's son. In fact, a few patients with "unrelated parents" stated that they would not know of a distant relationship, if there was any, between their parents.

Two marriages in Table I are surprising. None of the three communities, at least in its explicit tradition, permits the marriage of a woman with her paternal uncle. Yet, one has been recorded in the *Harijan* marriages. Islam and Christianity forbid the marriage of a woman with her maternal uncle, although a grudging dispensation to marry either uncle may be obtained from the Catholic Church with difficulty at least in Europe and America. It is surprising that we came across a Christian uncle-niece marriage in the present sample. The couple were apparently recent converts to Christianity from Hinduism, as the patient stated their caste as *Sale*, a weavers' group among the original *Sudras*.

The mean coefficient of inbreeding for all the marriages in this sample is found to be 0.0228 ± 0.0018. The percentage of first cousin marriages is significantly higher than other kinds of consanguineous marriages. All the first cousin marriages are between children of a brother and sister, but never of two brothers or two sisters. The latter type are permitted in Muslim and Christian communities but not among Hindus. But first cousin marriages of the type of a boy marrying his maternal uncle's daughter are much more in number than those who marry their paternal aunt's daughter. The former type were known to us to be preferred by many people of the state.

No significant correlation was found between the frequency of consanguineous marriages and illiteracy or rural background, though the latter two were correlated.

Discussion

It is clear from the data presented that there has been a very high rate of inbreeding among the people of Andhra Pradesh. This has been known and socially accepted for many hundreds of years.

Dube[1] gives a brief account of consanguineous marriages in the village of Shamirpet 25 miles from Hyderabad which is now the capital of Andhra Pradesh. He writes, ". . . out of the 340 Hindu marriages analysed by us, only 18 percent were cross-cousin marriages, the rest being negotiated marriages into unrelated families. Muslims permit both parallel and cross-cousin marriages, and indeed regard them as highly desirable. Our sample of Muslim marriages is not quite adequate, but out of 40 marriages fully investigated 19 were between cousins." It is noticeable that Dube[1] and his co-workers did not come across any uncle-niece marriages among Hindus. This may be because of the different way in which the two samples were delimited, i.e., the difference between a complete village and the inmates of two highly specialized wards. The products of uncle-niece marriages may be concentrated in maternity and gynecology wards, either because they are slightly abnormal in reproductive function, or as fetuses cause difficult pregnancies. This explanation of the difference would be very likely in a population where consanguineous marriages are rare. Alternatively, because Shamirpet had been within the former Muslim state of Hyderabad for over three centuries it is likely that the customs of its Hindu inhabitants have come to approximate in some respects those of their Muslim neighbors and thus to differ from those in other Telugu-speaking areas. The higher frequency of first cousin marriages in Dube's sample compared with our own

may be a compensation for the dropping of the uncle-niece marriage.

Sanghvi *et al.*[4] found lower frequencies of consanguineous marriages in 12 endogamous groups in Bombay. Many Marathi-speaking Hindus have a preference for a marriage between a boy and his maternal uncle's daughter, as do members of the present sample, but Sanghvi *et al.* do not mention any uncle-niece marriages.

The frequency of recessive allelomorphs in a population with a long established mating system is expected to be in equilibrium, and this equilibrium in populations like the present, with a high coefficient of inbreeding, will differ from those, like the European populations, with a low coefficient. Therefore, in Andhra Pradesh we should not expect to find as great an increase in the frequency of abnormal offspring among the progeny of consanguineous unions as is observed in Europe. The main purpose of our study is to collect data on this point. To be able to collect such data we are using the acknowledgedly biased sample of a hospital population. In such a sample the patients will more readily accept medical examination and we can consult their medical histories. The study is in progress and no conclusive statement about the genetic effects of inbreeding can be made at this stage.

Summary

Out of 327 marriages in Andhra Pradesh, 102 were consanguineous, including 29 between uncle and niece. The mean coefficient of inbreeding is found to be 0.0228 ± 0.0018.

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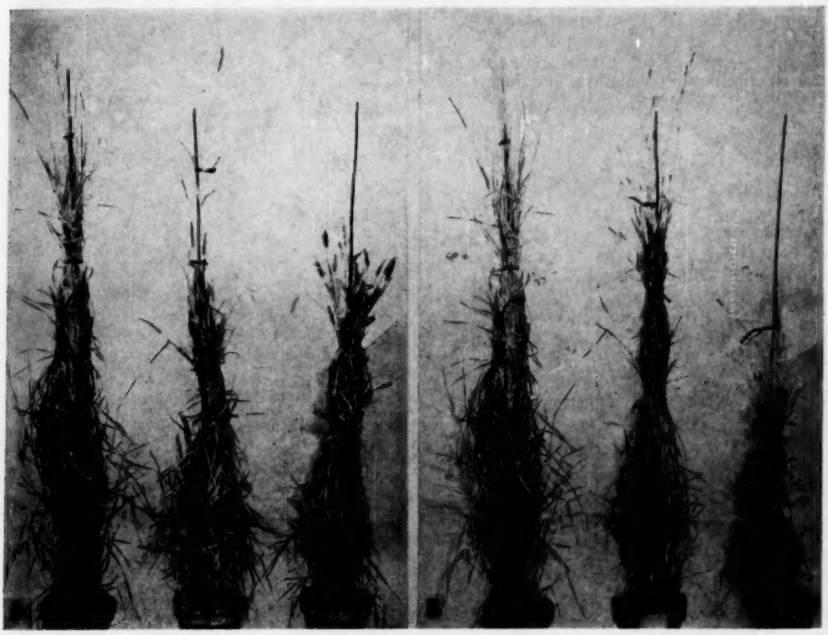
WILD BARLEY HYBRIDS

II. *Hordeum marinum* × *Hordeum compressum*

A. B. SCHOOLER*



PARENT AND HYBRID PLANTS

Figure 2

A—From left to right are shown *H. compressum*, F_2 or allotetraploid, and *H. marinum* (No. 43). *B*—*H. compressum*, F_2 or allotetraploid, and *H. marinum* (No. 56-39).

SCHOOLER[6] suggested in a previous paper that interspecific hybrids of *Hordeum* may cross more readily with cultivated barley than may the parents of the hybrids. Therefore, distinct lines of the wild species *H. marinum* were crossed with *H. compressum*, another wild species. Attempts to cross the two lines of *H. marinum* with *H. vulgare* have not been successful. (Other research workers[1-5] have reported crossing studies among wild and cultivated *Hordeum* species and other genera.)

This study is primarily concerned with the transfer of genes conditioning spot blotch resistance found in the wild *Hordeum* species to cultivated barley. One of the lines of *H. marinum*, No. 56-39, was found highly resistant, and No. 43 moderately resistant to the disease. *H.*

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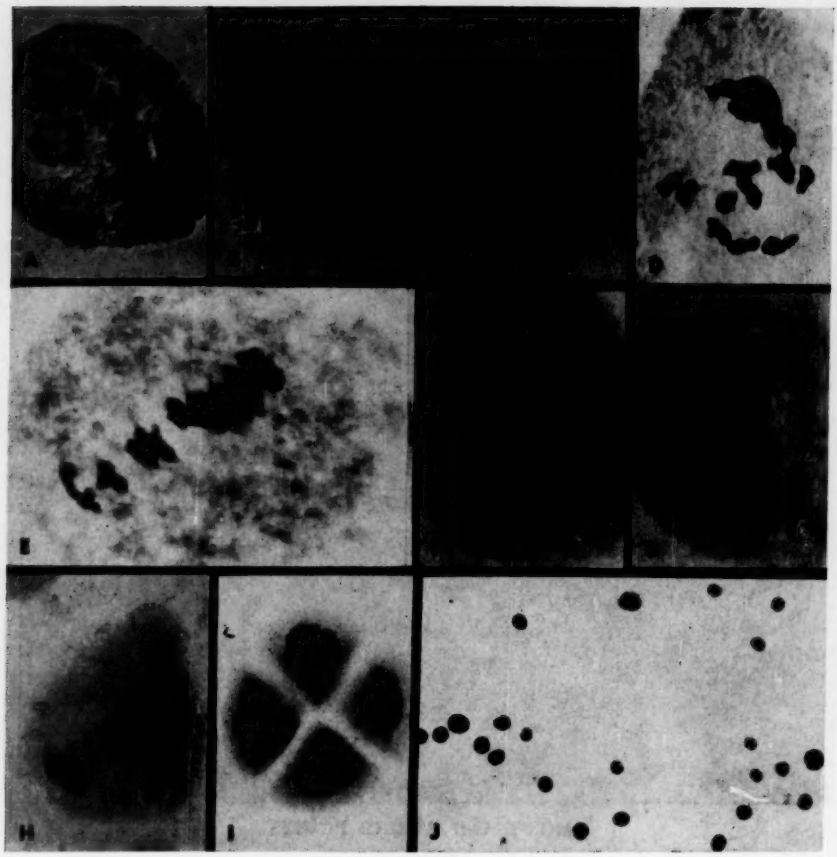


CHROMOSOME BEHAVIOR OF *H. MARINUM* (NO. 43) × *H. COMPRESSUM*

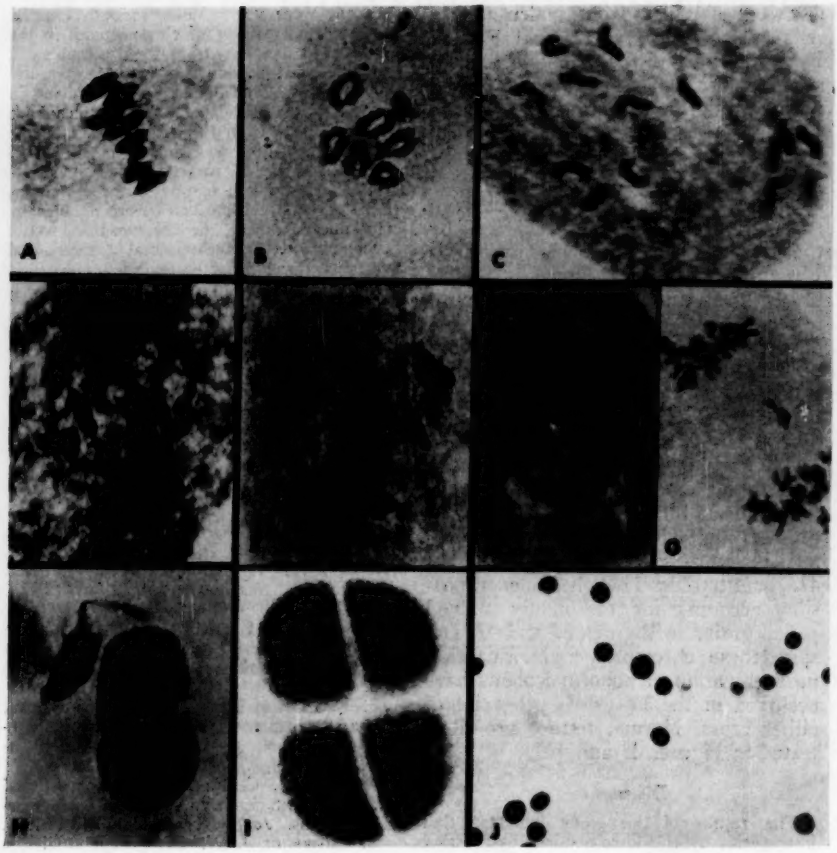
Figure 3

A—*H. marinum* (No. 43). *B*—*H. compressum*. *C*—Chromosome behavior of F_1 plants. *D*—Diakinesis, F_2. *E*—Metaphase, F_2. *F*—Anaphase, F_2. *G*—Anaphase, F_2. *H*—Dyad with chromosome fragment, F_2. *I*—Tetrad, F_2. *J*—Pollen, F_2.

compressum (No. 56-31) is highly resistant but is not immune to attack by the spot blotch organism.

Materials and Methods

Lines of *H. marinum* (No. 56-39 and No. 43) and *H. compressum* (No. 56-31) were obtained from Dr. G. A. Wiebe, U. S. Department of Agriculture, Beltsville, Maryland (Figure 2). Each of the two lines of *H. marinum* were crossed with *H. compressum* in the greenhouse during the winter of 1958. The interspecific hybrids, obtained through the use of the embryo culture technique[7], were sterile. However, seed was obtained from the F_1 hybrids after colchicine treatment, administered by



CYTOLOGY OF *H. MARINUM* (NO. 56-39) × *H. COMPRESSUM*

Figure 4

A—*H. marinum* (No. 56-39). *B*—*H. compressum*. *C*—Chromosome behavior of F_1 plants. *D*—Diakinesis, F_2. *E*—Metaphase, F_2. *F*—Anaphase, F_2. *G*—Dyad with chromosome fragment, F_2. *H*—Dyad showing micronuclei, F_2. *I*—Tetrad, F_2. *J*—Pollen grains, F_2.

submerging the crowns and roots in a .05 percent colchicine solution.

Cytological studies were conducted on pollen-mother-cells to determine the chromosome number and behavior of the parents, untreated F_1 plants, and the F_2 plants derived from seed of colchicine-treated hybrid plants. Pollen maturity was studied by use of the potassium iodide technique through starch formation. Normal pollen grains generally stain a dark blue by this method.

Results

Seed and spikes of the F_2 plants were larger, and generally the F_2 plants were slightly taller than the parents (Figure 2). No segregation was noted in the F_2 progenies. The F_2 plants were very similar in appearance to the F_1 but were highly fertile.

Chromosome counts and behavior stud-

ies were obtained from pollen-mother cells of the parents, untreated F_1 and F_2 plants. Each of the parents contained seven chromosome pairs that behaved normally during the meiotic process (Figures 3*A* and *B*, 4*A* and *B*). In each cross, chromosome behavior of the F_1 plants during meiosis indicated a nearly complete absence of homology between parental chromosomes, as evidenced by the lack of pairing during diakinesis (Figures 3*C* and 4*C*). However, the F_2 plants from treated F_1 plants showed nearly normal pairing and disjunction of chromosomes during meiosis (Figures 3*D-G* and 4*D-F*). These results suggested that the pairing in F_2 was between parental genomes. Although the diakinesis and anaphase movements are highly regular in the above figures, a few chromosome abnormalities such as fragments and bridges were noted (Figures 3*H* and 4*G*). In some cases micronuclei were observed in dyads (Figure 4*H*) and tetrads. This phenomenon possibly accounts for the lightly stained pollen grains in Figures 3*J* and 4*J*. Despite these chromosome abnormalities, no segregation or abnormal plants have occurred in the F_2 or F_3 progenies of either cross. Normal tetrads are illustrated in Figures 3*I* and 4*I*.

Discussion

The results of this study indicated that interspecific hybrids were obtained by crossing two lines of *H. marinum* with *H. compressum*. The almost complete absence of chromosome pairing during meiosis of the F_1 plants can be explained by the apparent nonhomology of the parental chromosomes. This phenomenon also revealed the similarity of chromosome behavior and homology that is expected in lines of the same species, as the chromosome reactions were very similar in both crosses.

The normal pairing of chromosomes in the F_2, which had a $4n$ chromosome complement, suggested that pairing was between parental chromosomes. The diploid type of pairing in the F_2 further indicated that allotetraploids were produced from these crosses.

The absence of segregation in the F_2 and F_3 progenies of these crosses provided evidence of the allotetraploid nature of the F_2 plants. If the parental chromosomes were doubled by colchicine treatment, the chromosomes would be homologous for the parental genomes and segregation would not be expected.

Summary

Allotetraploid plants were produced by doubling the chromosomes of the interspecific F_1 hybrid plants from crosses of two lines of the wild species *H. marinum* with *H. compressum*. Chromosome doubling was initiated by submerging the roots and crowns of the above plants in a .05 percent colchicine solution. Subsequent generations of these plants were highly fertile.

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FASHION IN THE COLOR OF SHETLAND PONIES AND ITS GENETIC BASIS

W. E. CASTLE*

THE Shetland pony came to North America in the eighties and nineties of the last century as a plaything for children or for parents who had not quite grown up and had surplus stable room and unused pasturage for animals.

The commonest colors in the animals imported from England or directly from the Shetland Islands were bay, brown and black, in a ratio of about 1:1:2. These three groups of black-pigmented animals differ from each other in possessing each a distinctive allele of the basic pattern gene, bay[1].

This pattern finds fullest expression in bays which have the genotype *AB*. It has only partial expression in browns, which commonly show light areas only on nose or flank. According to Odriozola[3], the Spanish geneticist, it has a less effective allele of *A*, viz., a^t, comparable to the black-and-tan allele of gray in the rabbit. Therefore, its genotype is a^tB.

In blacks the *A* gene is absent or ineffective, which is expressed in the genotype *aB*.

Among the imported animals were also found genuine brown-pigmented ponies of genotype *bb*, known as chestnut or sorrel. This genotype, recessive to all black-pigmented types (bay, brown, and black), was and still is well known in other breeds of horses. Its genuineness among Shetland ponies is attested by the frequent notation in sorrels, "with light mane and tail." About 10 percent of the imported ponies belong in this category.

TABLE I. Relative size of color classes in successive periods

No.	Year	Bay	%	Brown	%	Black	%	Chestnut-Sorrel	%	Mouse, Dun, Buckskin	%
886	1880	188	21	195	22	382	43	90	10	31	3.5
922	1890	163	17	156	16	464	50	108	11.5	31	3.3
537	1907	70	13	142	26	260	50	59	11	6	1.1
277	1915	42	15	54	20	136	50	43	16	2	0.7
207	1932	14	7	10	5	87	41	Ch So SD 17 18 55 90	42	6	3.0
162	1950	8	5	5	3	39	24	Ch So SD P 20 18 67 4 109	67	1	0.6
200	1959	9	4.5	6	3	24	12	Ch So SD P 18 71 64 7 160	80	1	0.5

*421 Spruce Street, Berkeley, California. I am indebted to the officers of the American Shetland Pony Club for making available to me the data on pony populations included in Table I.

There was also found in the foundation stock about three percent of ponies which were carriers of the *D* (dilution) gene. These are described as mouse, dun, or buckskin in color.

All the genetic constituents of a palomino were accordingly present in the imported foundation stock, including *A, bb* and *D*. Indeed, the synthesis of such a genotype is occasionally recorded, as in the gold and white stallion, Rowland Prince Larigo, 27035, born in 1946.

It is possible, however, that at this time palominos were already being produced among Shetland ponies of two slightly different genotypes, one involving true chestnut (*bb*) plus *A* and *D*, i.e., *AbbD*; the other involving the new mutant type of chestnut, really a modified black through the interaction of gene *S* with gene *B*, resulting in the genotype *ABS*. It is impossible to state to which type Rowland Prince Larigo, 27035, belonged, since among his ancestors were carriers of the mutant gene *S* (silver).

Less uncertainty exists in the case of the mare Blondy May, 18050, described as cream with white mane and tail, born in 1915. All her ancestors, both paternal and maternal, trace back to ponies born before the birth of Chestnut in 1897, when the *S* gene first came to notice.

The incidence of this gene *S* was the truly revolutionary event in Shetland colors in North America. It probably originated by mutation in the mare Trot, 31, described as fawn in color, with white mane and tail. Her son Chestnut, sired by an intense black stallion, Prince of Wales (probably a dominant black), was the founding sire of all silver dapples, which became the prevailing genotype from 1930 on[2].

Table I shows that the original color types persisted in about their original proportions through 1915, but in the next 20 years there was an astonishing increase of chestnut and sorrel classes, which included the new mutant groups of genotype *BS*.

The 1932 population sampled in Table I includes 17 chestnut, 18 sorrel, and no less than 55 silver dapple of unmistakable genotype *BS*. This includes nearly half (42 percent) of the population sampled at that time, and this predominance of *S* genotypes persists in the subsequently sampled populations of 1950 and 1959.

It is interesting to note that palominos (P in population samples for 1950 and 1959) are gaining in recognition as a distinct subgroup, though probably of mixed character, a minority of the genotype common in horses *AbbD*, but a majority of the newer genotype *ABS*.

The surprising increase in the subgroup sorrel between 1950 and 1959 is to be attributed to a growing preference among breeders of the new mutant genotype, *BS*, for a clear body color, which is found in the sorrel (*ee*) but not in the silver dapple (*E*).

Summary

An examination has been made of the colors of ponies recorded in the Shetland Pony Stud Book in successive periods from the first introduction of Shetland ponies into North America, about 1880, down to 1959. The population samples examined are large enough to give a correct idea of what were the prevailing color classes in each period and what was the probable genetic constitution of each class.

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GENETIC STUDIES ON COLD HARDINESS IN MEDICAGO SATIVA L.

H. DADAY AND C. G. GREENHAM*

IN SPITE of the great importance of cold hardiness in *Medicago sativa*, very little genetic information has been published. Peltier and Tysdal[9] found that inbred lines showed a distribution indicating polygenic inheritance. Dexter[2] has extensively reviewed physiological and agronomical features of cold hardiness in *M. sativa*.

A common viewpoint is that cold hardiness is often associated with dormancy, so that increased cold hardiness may be accompanied by decreased winter growth. The review of Levitt[7] confirms that in general the rate of growth in autumn is inversely related to frost hardiness, particularly in annuals. On the other hand the results of Tysdal[10] show that extended day length, although not as effective as shortened day length in increasing hardiness, increased both hardiness and winter growth. However, he did not investigate the genetic relationship between the two variables.

Greenham and Daday[4] have reported that, after cold treatment, in both F_1 and F_2 generations of *M. sativa* there are significant correlations between low frequency resistance measurements and cold hardiness as determined by subsequent recovery, so that such measurements may be used as a quantitative criterion of cold hardiness. This paper investigates the inheritance of electrical resistance in *M. sativa*, and the relationship between electrical resistance and winter growth.

Materials and Methods

The following varieties of *M. sativa* were used in the investigations, which were done at Canberra, Australian Capital Territory:

African, originated in South Africa, winter growing under Canberra conditions, cold susceptible.

Hairy Peruvian, originated in South America, growing under moderate cold and short day conditions, cold susceptible.

Hunter River, assumed to be derived from the French Provence and adapted to parts of the Australian environment, moderate winter yielding and cold susceptible.

Ladak, introduced from North India, winter dormant and cold hardy.

Rambler, bred in Canada from Ladak, and *M. falcata*[6], creeping rooted, winter dormant and extremely cold hardy.

The above assessments of hardiness are based on the known behavior of the varieties, and on the determinations made previously[3] and in the present investigations.

Single plants, initially grown in the glasshouse, were exposed to natural hardening treatment[7] in the open. In winter they were removed from the soil, trimmed, treated in a modified deep-freeze unit under reproducible conditions, then brought to room temperature. Four measurements of electric resistance at low frequency (1000 c/s) were made with metallic probes on each crown, the mean resistance measurement in kilohms (1K = 1000 ohms) being taken for each plant. The same cold treatment was not used in each ex-

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periment, because the relative hardiness of a plant depends on both genetic constitution and age. As far as could be determined beforehand with small samples, the cold treatment selected for any experiment would allow the majority of the treated plants to recover.

A previous paper[4] has described the apparatus used and ratings for recovery, and has shown that there is a phenotypic correlation between electric resistance and recovery, the correlation coefficient being of the order of 0.5 to 0.6. In view of this correlation coefficient and those given in Table II, the electric resistance of the treated plants is used in the present paper as the measure of cold hardiness. By this procedure quantitative measurements may be rapidly made on large numbers of plants.

Results

Variation within Hunter River

Three propagules of each of eight randomly selected Hunter River clones were tested in July 1957, the cold treatment being six hours at −5°C.

TABLE I. Analysis of variance of cold hardiness (as electric resistance) between clones of Hunter River

Source	D.F.	Mean Square (kilohms)	E (M.S.)
Between clones	7	35.18*	$\sigma^2_e + 3\sigma^2_g$
Within clones	16	7.97	σ^2_e

$$\text{Heritability} = \frac{\sigma^2_g}{\sigma^2_g + \sigma^2_e} \qquad H = 0.53 \pm 0.23$$

*$P < 0.01$.

The analysis of variance of the figures obtained is given in Table I.

Differences between clones are highly significant. The heritability in a broad sense is 0.53 ± 0.23, suggesting that potential genetic variation exists within the variety.

General and specific combining ability experiment

The experimental material was comprised of 82 adult field plants of Hairy Peruvian, Hunter River, Ladak and Rambler varieties and their six F_1 diallel progeny lines (excluding reciprocal crosses). Seedlings were planted in the field in the spring of 1955 at 5 × 5 links spacing. The plants were tested in July 1957, the cold treatment being 10 hours at −8°C. After being measured for electric resistance, they were planted in the glasshouse, then assessed for recovery five weeks later. Angular transformation of the recovery ratings (0-6) was used, as before[4]. The results from the F_1 lines were subjected to analysis for general and specific combining ability, according to Method 4 Model 1 of Griffing[5]. The analysis is based on the assumption that the experimental material is a selected sample of *M. sativa* varieties.

The analyses of variance and covariance (Table II) show that highly significant differences exist between parental lines and F_1 crosses as regards electric resistance and recovery ratings. The correlation between electric resistance and recovery is high.

TABLE II. Analyses of variance and covariance of electric resistance values and recovery in *M. sativa*

Source	D.F.	Mean Squares: Electric Resistance (kilohms)	Mean Squares: Recovery (angular transformation)	Mean cross-products	Correlation
Parents					
Lines	3	1,328*	5,152*	2,606	1.02
Residual	29	48	287	61	0.52
Total	32				
F_1 crosses					
Lines	5	480*	2,750*	1,036	0.97
Residual	44	56	461	81	0.51
Total	49				

*$P < 0.001$.

The between-line correlation coefficients of 1.02 and 0.97 were calculated from the components of variance, and values so estimated may be greater than 1.0. These statistics indicate the relationship between electrical resistance and recovery for the means of infinite populations of each line, errors of measurement and nongenetic elements having been removed by the analysis.

The *within-lines* correlation coefficients (0.52 and 0.51), which are smaller than the *between-lines* correlation coefficients, are the result of both genetic and nongenetic effects, and are therefore difficult to interpret.

The mean electric resistance values of the F_1 crosses are given in Table III. The lowest value is given by the Hairy Peruvian-Hunter River crosses, and the highest value by the Rambler-Ladak crosses. Crosses of cold susceptible and hardy varieties give values between these.

The analysis for general and specific combining ability is given in Table IV.

It is seen that the mean square for general combining ability is highly significant, but that for specific combining ability is small and not significant. These results indicate that differences in combining ability of these varieties are additive; that is, there are no indications that values of F_1 are likely to be other than approximately equal to the mean of the parents.

Table V presents estimates of the general combining ability effects for cold hardiness, Rambler contributing a high degree of hardiness, in contrast with Hairy Peruvian which confers a low degree of hardiness on its crosses.

TABLE III. Mean cold hardiness of F_1 generations measured after cold treatment in terms of electric resistance (kilohms)

Varieties	Hunter River	Rambler	Ladak
Hairy Peruvian	9.5	17.7	17.3
Hunter River		25.1	22.5
Rambler			29.6

S.E. = 2.59.

Relationship between cold hardiness and winter growth

The association between cold hardiness and winter dormancy in ecotypes of *M. sativa* could result from past selection in cold climates where both hardiness and winter dormancy would be advantageous. The association could also arise from pleiotropic effects, that is, a physiological correlation.

The relationship between cold hardiness and winter growth was examined in an F_2, comprising 142 six-month-old plants derived from African × Rambler crosses. The plants, in pots, were moved to the open in early April, 1959, when they were cut back. They were harvested for winter yield in July, and then the crowns were tested for cold hardiness (cold treatment six hours at −8°C).

Individual dry matter yields are plotted against electric resistance values in Figure 5. Although there is wide distribution of points in the middle region of the figure, the correlation coefficient is negative ($r = -.38$). An unequivocal interpretation of this correlation co-

TABLE IV. General and specific combining ability analysis for cold hardiness (as electric resistance)

Source	D.F.	Mean Square (kilohms)
General combining ability	3	81.31*
Specific combining ability	2	1.17
Residual (calculated)†	44	7.75

*$P < 0.001$.

†$\sigma^2 = M_e/K_e$ where M_e = Error Component on a single plant basis, K_e = 8.29, the mean number per line.

TABLE V. Estimation of general combining ability effects for cold hardiness

Varieties	General Combining Ability. Effects	Order
Hairy Peruvian	−16.35	4
Hunter River	− 3.75	3
Ladak	8.55	2
Rambler	11.55	1
S.E. (g_1-g_1)	1.61	

efficient is not possible because it could be the result of at least four factors:

(a) Linkage. The characters winter production and resistance are in "repulsion" in the parents. Since only one segregation occurred in the production of this material, and since both characters are probably determined by many genes, some residual linkage may be expected.

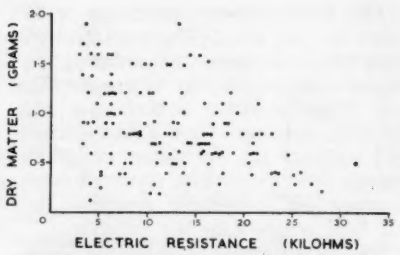
(b) Environmental effects. It is possible that local environmental variations could have correlated effects on the two characters.

(c) Experimental error in determining the characters. If the correlation coefficient were high, experimental errors would diminish its absolute value. Thus the assessment of yield might be complicated by factors such as seed size. However, differences of the order observed here most likely reflect true differences in growth rate in addition to nongenetic influence.

(d) Pleiotropic effects of genes.

A clarification of the relationship between the two characters and a reduction in the experimental errors was achieved by a different approach.

If an environmental treatment can be imposed to change one character, but not the other, this would indicate a degree of independence, and hence suggest the possibility of breeding genotypes showing opposite combinations of the characters; on the other hand if one character could not be altered without changing the other, this would indicate a degree of physiological dependence. As the growth of *M. sativa* in winter can be increased by extended day length[10], the following experiment was designed to test whether such a change in growth was accompanied by a change in cold hardiness.



CORRELATION BETWEEN COLD HARDINESS AND WINTER YIELD

Figure 5

Dry matter yields of individual F_2 plants are plotted against electric resistance values. Although there is a wide distribution of points in the middle of the figure, the correlation coefficient is negative (−0.38).

Adult field plants of randomly selected Hunter River, Rambler, and some selected winter growing F_1 hybrids of

TABLE VI. Effect of extended light on the cold hardiness (electrical resistance) and winter growth of *M. sativa*

Lines	Mean electric resistance (kilohms)		Mean log dry matter (gms)	
	18 hrs. light treatment	Natural daylight	18 hrs. light treatment	Natural daylight
Hunter River	17.1	9.2	0.55	0.28
F_1	14.6	12.8	1.22	0.40
Rambler	37.6	35.2	0.87	0.00

TABLE VII. Analysis of variance of cold hardiness (electrical resistance) and winter growth of *M. sativa*

Source of variance	D.F.	Mean Squares	
		Electric resistance (kilohms)	Log D.M. (gms)
Lines	2	6700*	1.89†
Treatment	1	448‡	12.15*
Lines × treatment	2	117	1.05‡
Clones	54	175†	0.43
Residual	54	87	0.29

*$P < 0.001$.
†$P < 0.01$.
‡$P < 0.05$.

susceptible and hardy parents were divided into two clonal propagules, one for each treatment. They were planted in seven-inch pots at the end of May 1957 and randomized in two areas in the open. The plants in one area were exposed to natural light, and those in the other area were subjected to a day length of 18 hours, obtained by means of 200-watt filament lamps six feet above the plants (the lamps operated only at night and gave a mean intensity of 27 foot-candles at plant level). Harvesting and testing were done in early August 1957, the cold treatment being six hours at $-7°C$. The tops of the plants were used for dry matter determinations.

Tables VI and VII give the mean values for electric resistance and winter growth of the lines under natural and extended daylight. The analysis of variance indicates that there are significant differences between lines for both electric resistance and winter yield. The electric resistance value of Hunter River was nearly doubled by extended light whereas that of other lines was only slightly affected. However, all this response in Hunter River occurred in dormant plants ($P < .001$) and the resistance of nondormant plants was scarcely affected. While we are unable to explain the significance of this phenomenon, we consider it should be mentioned here. Over all lines, extended light increased both electric resistance and winter yield significantly.

Thus in the lines investigated the increased winter growth resulting from the treatment did not adversely affect hardiness. The line $\times$ treatment interaction was significant for winter growth only, which indicates that the lines differ in respect to treatment response. The clonal differences were significant for cold hardiness but not for winter growth.

The F_1 plants were a sample selected for growth rate and thus could not be used to estimate the genotypic (between clone) correlation. Separate analyses of covariance are presented for the two varieties in Table VIII. Estimates of the residual correlation are -0.19 and -0.05 in the two varieties. These suggest that responses by a genotype in yield were practically independent of responses in resistance. But the low values could also reflect errors of measurement and environmental correlations so that these results cannot be interpreted unequivocally. The between-clone correlations are -1.1 and -0.72. These results must obviously be interpreted with caution, but they do suggest that there is a strong negative genotypic correlation between winter growth and electrical resistance values. Such a possibility is further suggested by the value for the F_1 (Table VI). This F_1 population was obtained by selecting winter-active plants from a number of F_1 lines from crosses involving six different varieties. The weighted mean resistance index of the parents would be approximately 20. From the diallel analysis one would expect the F_1 to have approximately this value, whereas the observed

TABLE VIII. Analysis of variance and covariance of cold hardiness (as electrical resistance) and winter growth of Hunter River and Rambler varieties.

Source of variance	D.F.	Mean Squares: Electric resistance (kilohms)	Mean Squares: Log D.M. (gms)	Cross products
Hunter River				
Treatments	1	585.80†	0.6979	20.22
Clones	18	109.02	0.4196	−4.02
Treatments × clones	18	65.92	0.2259	−0.74
Rambler				
Treatments	1	53.00	6.43*	19.53
Clones	18	267.11	0.30	−4.90
Treatments × clones	18	108.94	0.28	−0.24

*$P < 0.001$.
†$P < 0.01$.

value was 13. The discrepancy probably results from a correlated response of electric resistance to selection for winter growth.

However, the fact that an environmental treatment (extended day length) increased both winter growth and resistance values, and the way in which the F_2 values are distributed in Figure 5, give some cause to hope that recombination of the two variables may be possible.

Cold hardiness of the leaves, which could be relevant to fodder digestibility, has also been investigated. The correlation coefficient between yield and percentage of uninjured leaves is small ($r = 0.15$) and not significant, indicating that high winter yield can probably be combined with freedom from frost damage to the leaves.

Discussion and Conclusions

The present investigations with *M. sativa* demonstrate clear differences within and between varieties in cold hardiness and in associated general combining ability. The F_1 generation is intermediate in hardiness between its parents and there are no indications of nonadditive genotypic variance. The results suggest that F_1 values may be predicted from the parental values, and that genotypic differences in an F_2 generation might be largely additive. This result does not exclude the possibility that nonadditive effects between other varietal crosses might occur or that nonadditive genotypic variance might not exist in a random breeding population. Moreover, the nature and extent of variation in an open pollinated variety or in varieties in which selection is practiced cannot be inferred from this study.

The segregation in an F_2 generation of widely different parents suggests that recombination of cold hardiness and winter growth might occur, and that selection for both might be practicable. The increased growth caused by extended light was accompanied by an increased winter hardiness as measured electrically, and supports the above conclusion. However, selection for recombinants might be seriously hampered by the negative genotypic correlation.

Improved hardiness as a result of crossing and/or selection from a large population can be simplified (1) by using seedlings instead of adult plants[8], (2) by a preliminary screening of the cold-treated seedlings on the basis of electric resistance, and (3) by a final screening on the basis of rate of recovery of the selected plants[4].

The above investigations were carried out under comparatively mild winter conditions, so that varietal differences were not as marked as would be expected in more severe climates. Moreover, the cold treatments applied were intentionally not very severe. Bula *et al.*[1] indicate that degree of hardiness depends *inter alia* on environmental conditions. The results of the present investigation may not be applicable to areas where winters are very severe.

The economic importance of winter production of *M. sativa* under comparatively mild winter conditions suggests further investigation of problems basic to cold hardiness and winter growth and encourages an intensive search for useful recombinants. The exact degree of cold hardiness required, and the physiological relationship with other productive characters, remain subjects of great importance to Australian agriculture.

Summary

The cold hardiness of single plants of *M. sativa* was determined by means of electric resistance measurements after cold treatments. Differences were demonstrated between clones of the Hunter River variety, heritability in the broad sense being 0.53 ± 0.23.

From a study of crosses of Hairy Peruvian, Hunter River, Ladak and Rambler it is concluded that the average performance of these parents in crosses is significantly different, and that the F_1's are generally intermediate in hardiness between the parents. There are no indications of dominance or nonadditive genetic effects. Rambler contributed most to the hardiness of the F_1 lines.

In an F_2 between hardy and susceptible varieties, the correlation coefficient between electric resistance and winter yield was -0.38. The genotypic correlation within two varieties between the two variables was strongly negative. Extended light treatment increased both winter yield and electric resistance in a mixed population. These results suggest that recombinants with good winter growth and moderate hardiness might occur, but that selection might be seriously hampered by a negative genotypic correlation between these desirable characters.

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◆

THE CATTLE BREEDS OF THE U.S.S.R.

THE chief value of this book* lies in Part II which contains descriptions of the origin, characteristics and productivity of cattle breeds native to the U.S.S.R., of which twenty are described. Six indigenous breeds are the Kholmogor and Pechora (dairy types), the Caucasian (dual purpose), the Ukrainian Grey (triple purpose—meat, milk and work), and the Kalmyk and Kirgiz which are regarded primarily as beef animals. The Kholmogor has a small amount of Holstein-Friesian blood. The other breeds, which were produced from cross-bred foundations, using native cattle crossed with one, two or even as many as six or seven western European breeds, together with other native breeds in some cases, are the Aulie-Ata, Bestuzhev, Gorbatov Red, Istoben, Kostroma, Latvian Red, Siberian, Suksun, Tagil, Tambov, Ukrainian Red, Ukrainian Whitehead, Yaroslavl and Yurino. All are either dairy or dual-purpose in type, with little emphasis on beef qualities.

Descriptions are also given of twelve western European breeds that are used more or less extensively in the U.S.S.R., including data on their productivity; of these, the Simmenthal is most important; also, there are discussions of the use of the Zebu, Yak and Water Buffalo.

The initial chapters, which constitute Part I of the book, while shedding considerable light on the thinking of animal breeders in the U.S.S.R., contain little that is new, and are sprinkled generously with the thinking of Lysenko and Michurin. Fortunately, there is little evidence that their theories have adversely affected the material on breeds which, on the whole, appears to constitute an objective job of reporting.

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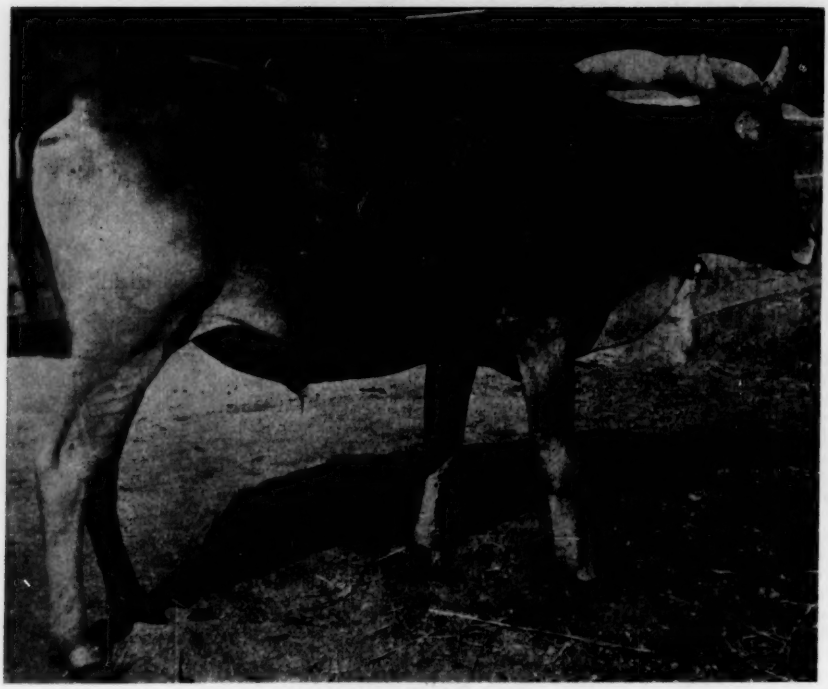
U.S. Department of Agriculture

**Breed Improvement in Cattle Breeding* by E. A. Novikov, D. I. Startsev and E. A. Arzumanyan. This book is not new. It was published in 1950 by the State Publishing House for Agricultural Literature in Moscow, the Russian title being *Plemennoe delo v skotovodstve.* However, the English version, issued for the National Science Foundation and the U.S. Department of Agriculture in 1960, is new. Translated by R. Farkash, it is available from the Office of Technical Services, U.S. Department of Commerce, Washington 25, D. C. $4.00.

THE SHORT-TAILED CATTLE OF THAILAND

DAVID C. RIFE*



SHORT-TAILED OX

Figure 6

The tail, including the switch, appears normal in all respects except length. Presumably the short tail is characterized by less than the normal number of caudal vertebrae. (Photographs were supplied by Dr. Ratana Oonygawongse.)

THE native cattle of Thailand are unusually hardy, small animals. They are not used for milk production, but are raised primarily for draft purposes, although some are slaughtered for beef. Mature cows average between 400 and 500 pounds in weight. They range in color from reddish yellow to black, the majority being of the lighter shades. They are long legged, have a slight hump, and possess larger dewlaps than western breeds.

The total cattle population of Thailand is estimated to be around 5,000,000 head. The Thai Department of Livestock Development employs castration teams which go throughout the country to select male calves to be used for

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SHORT-TAILED CALF

Figure 7

The dam of this animal has a normal tail; its sire is unknown. However, the sex ratio within the herd indicates that the trait is neither sex-linked nor sex-influenced.

breeding purposes and to castrate the rest of them. Those that are castrated develop into oxen which are used for plowing, hauling produce to market, and other draft purposes. Cattle are most abundant in relatively drier portions of the country, water buffalo being more commonly employed in the heavy rice growing areas of the central plains.

While I was serving as a Livestock Advisor in Thailand, a member of the Department of Livestock Development* mentioned that he remembered a herd of native cattle he had seen 30 years previously, many of which were characterized by short tails. Like many herds of cattle in remote and isolated parts of the country, there probably had been a great deal of inbreeding in it. Inbreeding, plus castration of good bull calves for draft purposes, may be partly responsible for the small size of the indigenous cattle.

The Department of Livestock Development in Thailand is interested in the improvement of the indigenous cattle. One way of going about this is hybridization of inbred lines. It was decided to find out whether or not the herd with the high frequency of short tails was still in existence. If so, it might provide breeding stock for an inbred line which could be hybridized with cattle from a different part of Thailand.

When further inquiry revealed that the herd still existed, Dr. Ratana Oonygawongse, Chief of the Division of Animal Husbandry, and I accordingly ar-

*Dr. Jones, Veterinary Advisor, Thai Department of Livestock Development.

ranged to take a trip to see these cattle. They are located in a mountainous jungle region in southeastern Thailand adjacent to the Thai-Laotian border, about 50 miles beyond the railhead at Ubon. The latter portion of the trip was made by jeep, and finally on horseback.

Figure 6 shows a short-tailed ox taken from the herd. The tail, including the switch, appears normal in all respects except length. Presumably the short tail is characterized by less than the normal number of caudal vertebrae. Figure 7 shows a calf from the same herd. Its dam has a normal tail, and the sire is unknown.

The herd exists in a semiwild state and depends upon grazing for its subsistence. Although we did not have an opportunity to see the entire herd, local veterinarians reported that it consists of 28 animals, of which 13 possess short tails. Among the latter, six are of one sex and seven are of the other, as nearly equal numbers as possible in the two sexes.

It was impossible to obtain information that could tell us whether short tail is inherited as a dominant or a recessive. The sex ratio indicates that it is neither sex-linked nor sex-influenced. Apparently there are just two phenotypic categories, either long or short tailed. This, plus the high incidence of short tails within the herd, strongly suggests a one-gene difference between long and short tails. This trait has not been reported in any other herds of cattle throughout Thailand.

The Division of Animal Husbandry plans to purchase a short-tailed bull for use in its breeding program. Thus it should be possible within a few years to ascertain how short tails in the cattle of Thailand are inherited.

◆

National Science Foundation Research Proposals

The Division of Biological and Medical Sciences of the National Science Foundation has announced that the next closing date for receipt of basic research proposals in the Life Sciences is May 15, 1961. Proposals received prior to that date will be reviewed at the summer meetings of the Foundation's advisory panels and disposition will be made approximately four months following the closing date. Proposals received after the May 15, 1961, closing date will be reviewed following the fall closing date of September 15, 1961. Inquiries should be addressed to the Biological and Medical Sciences Division, National Science Foundation, Washington 25, D. C.

COMPLEXITIES OF HUMAN TASTE VARIATION

GUNNAR SKUDE*

INDIVIDUAL taste variations for phenylthiourea (P.T.C.) and for creatine were observed in 1931 by Fox[8, 9] and by Williams[28]. Since then the interest of geneticists has been focused mainly on P.T.C. and other SCN compounds (review by Kalmus[15]). The observation by Blakeslee and Fox[3] that "the taste world of one individual is different from that of another" has obtained ample confirmation through subsequent studies. Quantitative and qualitative taste differences for various chemical compounds have been observed by several investigators and reviews have been given by Cameron[7] and Williams[30].

Inter-individual taste variations for a series of compounds have been observed among children and adults in southern Sweden by Skude[24, 25]. These differences are summarized in Table I.

It may be noted that the sum of different qualitative responses to chemical taste stimuli is larger than the number of examinees in all rows. This is due to the fact that taste quality depended upon the dilution. The deviating taste qualities were usually described for weaker concentrations; such statements about the most concentrated solutions used were predominantly given to the bitter substances allylthiourea and quinine sulfate. This writer experienced appreciable difficulties in getting clear-cut answers from some examinees and some compounds, especially the two just mentioned. Also, strychnine and P.T.C. in weaker concentrations were sometimes described as having a peculiar, indefinite taste that "could be" bitter or almost any taste.

A smaller series (unpublished) was tested with the following substances: sodium nitrate which as a rule was described as salty, occasionally as bitter or sweet; acetic acid which was described as tasteless or sour; mannose which was described as sweet, sweet and bitter, bitter or salty; maltose which was described as sweet, sweet and bitter, bitter, sour or salty, or as having no definite taste; and dextrose which was described by all the tasters as sweet. Blakeslee[2] reported different taste reactions for mannose and P.T.C. Blakeslee and Salmon[5] found two persons of 47 to taste quinine as sweet in subliminal solutions, subliminal as far as bitter taste was concerned. Richter and Clisby[22], studying P.T.C. taste reactions, stress the difference between the taste-difference threshold as compared with the solvent, and threshold for the taste specific to the substance being tested. Varying taste responses have also been found for strychnine sulfate, arbutin, aspirin, hydrochloric acid, sucrose, sodium chloride, potassium chloride, saccharine, cascara and others including P.T.C.[12, 16, 17, 18, 29]. A review can be found in Moncrieff[20].

Procedure for Elimination of Semantic Errors

Taste is a personal sense. A person will have difficulties in concealing a color blindness, but his taste discrimination will by no means be equally challenged in everyday life and occupational settings. It is even possible that private taste denominations may run in families. The latter source of error was not observed in a series of observations on sweet taste quality for P.T.C.[24]. It could, however, probably be eliminated by adoption of the following means, observed to be useful in the foregoing study:

1. The choice of probands from age groups and intellectual levels appropriate to the

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investigation.

2. The use of a battery of compounds, described by a vast majority of people as definitely salty, sour, sweet, and bitter.
3. The use of a sufficient number of blanks.

In the present investigation the reliability of the results was probably enhanced by the choice of sweet taste perception as definitely different from "strong" or "unpleasant." However, it was this writer's experience that people of normal mental aptitude had few difficulties in identifying a taste described as salty with the taste of sodium chloride, a sour taste with a hydrochloric acid dilution and, with notable exceptions, a bitter taste with a quinine sulfate solution. By using solutions of increasing strength it was always possible to get a clear-cut response free from semantic errors. All examinees stated at least one of the stronger solutions to taste sweet, sour, salty, or bitter. When the taste of P.T.C. was described as sweet, the taste was always approximately identified with at least one dilution of sucrose or dulcine.

Probands selected from suitable age and intelligence levels do not, of course, represent a random sample of the general population. When a population exhibits no appreciable ethnic stratification, the biases introduced by the selection of probands would hardly influence the possibility of discerning trends of intrafamilial clustering of a not too common taste deviation, like the sweet taste perception for P.T.C. While it is possible, by means of the mimical response, to observe if an infant or a low-grade mental defective can taste P.T.C. as unpleasant or not, it would be difficult to get a clear response regarding the possible sweetness of the substance.

Procedure for Elimination of Guesswork

1. Blind tests consisting of the solvent.
2. Adoption of an assortment procedure.

Soltan and Bracken[27], using the method with impregnated filter paper, had blinds of plain filter paper. In the foregoing investigation[24] distilled water was used in blind tests. In an investigation of the prevalence of sweet taste perception for P.T.C. on an independent sample[25] as well as in a later study[26], the assortment technique of Harris and Kalmus[12] was used.

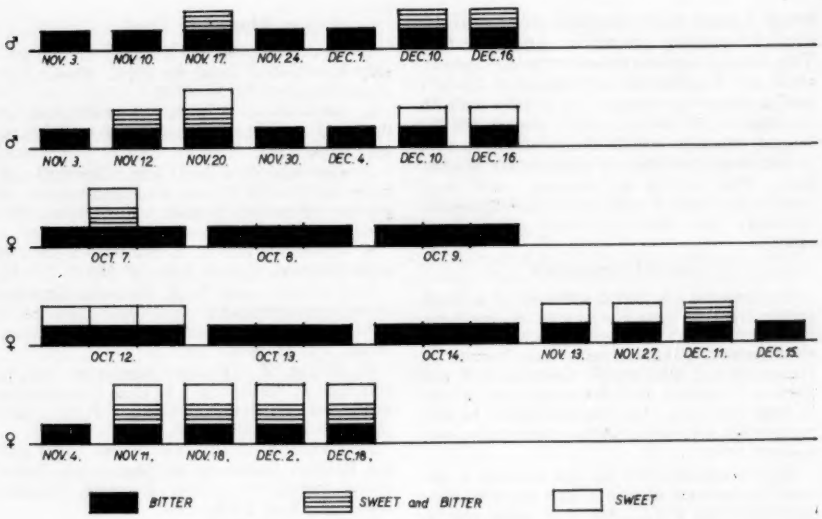
Procedure for Evaluation of Intra-Individual Taste Variation

Probands from the first family study of sweet taste perception for P.T.C. were reexamined at varying intervals for any intra-individual variation in taste quality[26]. The results are summarized in Figure 8.

None of the five persons whose taste responses are reproduced in the figure were consistent in their taste reactions. Variations in taste quality for P.T.C. sometimes occurred within a few hours. Salmon and Blakeslee[28] found variation in threshold concentration for P.T.C. within 15 minutes. Repeated examinations are necessary because of the intra-individual variations, which might be fairly common. No satisfactory explanation can be offered for this variation, but it probably depends on variations in the inner chemical combination of the individual[10, 14, 19, 21].

TABLE I. Taste qualities of seven different compounds

Compound and concentration	Predominant taste quality						Number of examinees
	Salty	Sour	Bitter	Sweet	Unclassified	Total	
Sodium chloride 0.04-0.80%	249	2	11	18	29	309	251
Hydrochloric acid 0.0015-0.0300 N	7	243	10	8	24	292	251
Allylthiourea 0.00059-0.01172%	4	5	136	47	62	254	184
Quinine sulfate 0.00005-0.00100%	2	3	226	48	47	326	251
Strychnine 0.0000004-0.0000080%	2	0	180	43	55	280	184
Sucrose 0.1-2.0%	0	3	21	251	19	294	251
Dulcine 0.00085-0.01700%	0	1	3	184	8	196	184


♂
NOV. 3. NOV. 10. NOV. 17. NOV. 24. DEC. 1. DEC. 10. DEC. 16.
♂
NOV. 3. NOV. 12. NOV. 20. NOV. 30. DEC. 4. DEC. 10. DEC. 16.
♀
OCT. 7. OCT. 8. OCT. 9.
♀
OCT. 12. OCT. 13. OCT. 14. NOV. 13. NOV. 27. DEC. 11. DEC. 15.
♀
NOV. 4. NOV. 11. NOV. 18. DEC. 2. DEC. 18.
BITTER
SWEET and BITTER
SWEET

TASTE REACTIONS FOR DIFFERENT P.T.C. CONCENTRATIONS

Figure 8

Probands from the first family study of sweet taste perception for P.T.C. were re-examined at varying intervals for any intra-individual variation in taste quality. None of these five persons showed consistency in taste reactions. Variations sometimes occurred within a few hours.

Methods Used in Taste Investigations

1. Crystals or powder.
2. Impregnated filter paper.
3. Solutions of different concentrations.

The intensity of the taste sensation produced by giving crystals or powder depends on the size of the grains. Hoyme[14] remarks that it is difficult to control the amount of crystals offered. This method has now been practically abandoned.

Impregnated filter papers have been used in several P.T.C. investigations. A disadvantage with this method is that it is not convenient for threshold determinations or for examination of the relative strength of taste sensations, or in investigations on how different taste qualities depend on concentration. Another disadvantage of this method is that the examinee might not place the strips of filter paper on the same place on the tongue. Hoover[13], using this method, found some persons to give unreproducible taste reactions for P.T.C., sodium benzoate, as well as for sodium chloride, citric acid, and quinine sulfate. Soltan and Bracken[27], using the same method, found inconsistent taste reactions for these compounds and to a minor extent also for sucrose. The results of this writer's experimental series support this behavior: sucrose gave more consistent taste responses than sodium chloride and quinine sulfate. Owing to the intra-individual taste variations and the variation of taste quality with the concentration it is necessary to use a test method taking these variations into consideration, i.e., the use of solutions of different concentrations.

Solutions given as drops on the tongue have the disadvantage that the drops, which probably vary in size, might not always be placed on the same area of the tongue. Salmon and Blakeslee[23] in determining P.T.C. threshold gave the examinee approximately 0.6 c.c. by the straw method[4]. They found this quantity to be sufficient, since there was no lowering of the threshold when the tasters were given larger amounts of a weaker concentration. Harris and Kalmus[12] recommend the following technique for investigation of taste thresholds. Successive solutions with halved concentrations are used. An approximate estimation of the threshold solution is made by using a single glass to compare with the solvent; the examinee is then given eight tumblers, four containing the roughly determined threshold solution and four containing water. The tumblers are randomly ordered and the person is told that four of the glasses contain water and four contain the substance. The examinee is asked to assort them in two groups of four according to their taste. If this procedure is correctly done, the test is repeated with weaker solution. The lowest concentration correctly as-

sorted is taken as the threshold solution. If desired, the tumblers are refilled during the test. This method appears to be exemplary; guesswork and imagination are eliminated. Hahn[11] used a "taste-microscope" (a U-tube with an opening at its lowest point, placed on the tongue) through which the solutions flowed, to determine the effect of temperature of solutions. This method is, however, very time-consuming. Bujas[6] used a modified apparatus to study the effects of two simultaneous stimuli.

General Comments

On variation of tasting capacity of a given person from one occasion to another, the cause might be sought in varying biochemical constitution and endocrine secretion. Results by Richmond and Abarbanel[21], Goetzl *et al.*[10], and Hoyme[14] support that hormones play a role in taste reactions. Age changes cannot be held responsible for the variations observed in such a short time.

Such inconsistencies do not exclude a genetic background of such a trait as sweet taste perception for P.T.C., but they make conclusions more difficult than would appear when family members are observed only once, as in the foregoing study. Then the conclusion that the observations were consistent with a dominant mode of inheritance of the sweet tasting ability for P.T.C. was formally correct, but has to be seen in the light of the additional disagreement between observations on a given occasion, or on two near-lying occasions, and a subsequent repetition of the examination after a longer interval and on repeated occasions. Because of the wide inter-individual variations that prevail with respect to taste, taste would be a fruitful field for genetic studies, except for the complications discussed here.

Summary

Inter- as well as intra-individual taste variations occur and might be due to inheritable characteristics. The intra-individual variation can occur within a short period, but it is probably limited, the limits varying from individual to individual.

To determine taste reactions of a given person, it is necessary to repeat the test. The taste quality is dependent on concentration. For this reason it is convenient to use solutions of different concentrations. A discriminating procedure, that implies separating of four tumblers containing a solution of the substance and four tumblers containing the solvent, reduces the guesswork and dangers from the play of imagination to practically nil in threshold determinations. Different concentrations of substances, representing the four primary tastes, must be included in the test to avoid semantic errors. Blind tests might still more increase the validity of the results.

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RIB FUSIONS, A NEW MUTATION IN THE MOUSE

JUDITH A. MACKENSEN AND LEROY C. STEVENS*

RIB FUSIONS is a new skeletal mutation in the mouse, which consistently causes fusion of the ribs. The mutation is a semidominant, autosomal gene with regular manifestation, and has been given the symbol *Rf*. Evidence will be presented indicating that the gene is lethal when homozygous. This new mutation has provided a useful tool for investigations of the role of gene action in the development of the ribs[6].

The original *Rf* mutant, a female with pronounced tail kinks, was found in a litter produced by a pair of strain 129/RrSv mice†. This pair produced 31 offspring, of which two females had kinky tails; the mutant colony is descended from one of these. The *Rf* mutation is maintained on two genetic backgrounds: the original 129, and descendants of crosses of C3HeB/FeJ females and 129 *Rfrf* males.

These colonies are continued by the matings of two kinky-tailed, or of kinky- and normal-tailed animals, usually sibs. Breeding pens are checked weekly; those with pregnant females are then checked daily (except Sundays) for new litters. Affected animals were tested for homozygosity at the *Rf* locus by mating them with normal, strain 129/Sv mice, and examining the offspring for the presence of rib fusions and/or tail kinks. Term embryos were obtained from normal- and kinky-tailed females mated with kinky-tailed males and killed 18 or 19 days after the appearance of the copulation plug. Skeletal preparations were made by clearing alcohol-fixed material in KOH and staining the bone with alizarin red.

Genetics

In the early phases of this study, animals were classified on the basis of kinky tails only. Skeletal study of cleared specimens revealed that affected animals are characterized by a high incidence of fused ribs as well as tail kinks. Accordingly, litters have been classified on the basis of the presence of fused ribs in cleared or fresh material. Animals which are kept for breeding purposes are classified by the presence of kinks in the tail, which usually do not appear until the animals are seven to 10 days of age.

The results of classification on the basis of fused ribs are presented in Table I. The data conform to that expected for a semidominant gene which is lethal in the homozygous state (*RfRf*). Fewer heterozygotes were obtained than expected, suggesting some pre- or postnatal loss of this class as well. The production of abnormal offspring by two normal-tailed parents indicates that some of the animals classified as normal on the basis of tail kinks only may also have fused ribs, and therefore be of the genotype *Rf-*. Males and females were affected in equal numbers.

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†Standard symbols for the inbred strains are in accordance with the rules and standard symbols given in "Standardized Nomenclature for Inbred Strains of Mice, 2nd Listing," prepared by the Committee on Standardized Genetic Nomenclature for Mice, published in *Cancer Research*, Vol. 20: Feb. 1960.

Kinky-tailed animals (*Rf*-) were tested wtih normal, strain 129 mice (*rfrf*) in order to determine whether they were homozygous or heterozygous for the gene *Rf*. All the offspring derived from these crosses were autopsied and examined for fused ribs and tail kinks. An animal being tested would be considered to be *Rfrf* if it produced five to 10 normals out of a total of 10 to 20 offspring. Thirty-two mice, 20 males and 12 females of both colonies, were tested this way and all were found to be of the genotype *Rfrf*. This evidence further indicates that the gene *Rf* may be lethal when homozygous.

Description

Litters segregating at the *Rf* locus cannot be classified easily at birth, unless autopsied, since most members have straight tails, and there are no other recognizable defects such as reduced size or neonatal anemia. Occasionally animals are born with shortened and/or kinky tails, but usually kinks develop by seven to 10 days of age in the *Rfrf* animals. In adults, the tails are kinked to a variable extent, and the bodies often appear somewhat shorter and wider in those with extreme kinks than they do in normal mice. At 60 days of age, *Rfrf* animals are slightly smaller than their normal-tailed sibs (Table II).

Skeletal Defects

Newborn and term embryos

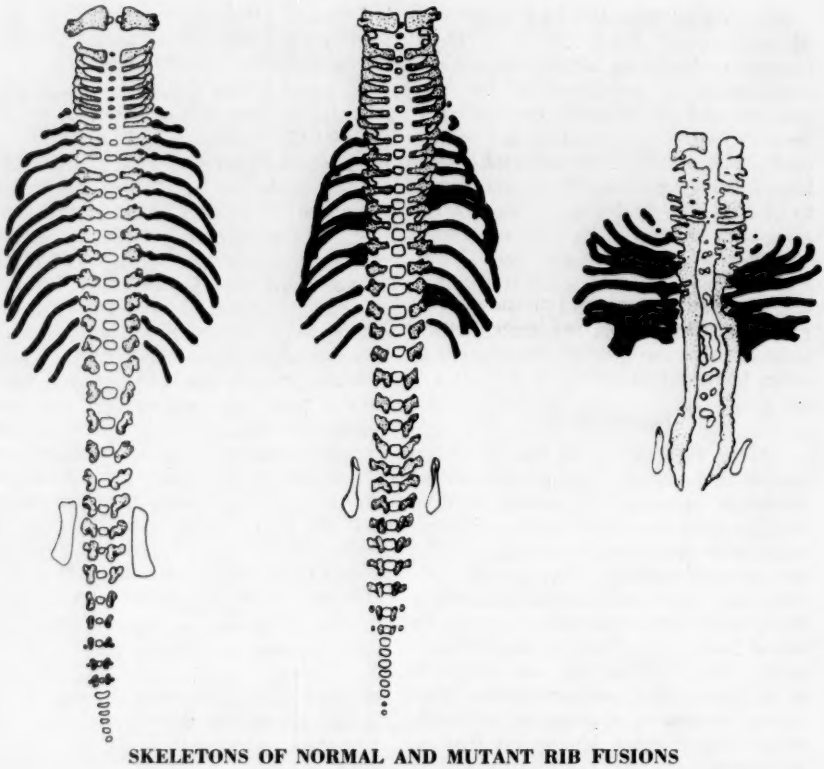
Complete litters of this age were cleared and classified as normal or abnormal skeletons. This group consisted of 139 abnormal skeletons, presumably *Rfrf*, and the normal littermates. The majority were from the two *Rf* colonies, but some were obtained from matings of *Rfrf* males with BALB/cJ, A/J, RF/J, and DBA/1J females. The genetic backgrounds did not appear to alter the expression of the *Rf* gene.

A study of the types of skeletal abnormalities showed that four percent of the 139 *Rfrf* skeletons had anomalies of the cervical, 32 percent of the thoracic, 26 percent of the lumbar, 16 percent of the sacral and 32 percent of the caudal vertebrae. The following types of abnormalities were observed: fusions of adjacent neural arches (44 percent), fusions of adjacent centra (11 percent), hemicentra (four percent), bilateral centers of ossification either completely (seven percent) or partially (24 percent) separated from each other, and thoracolumbar curvatures and displaced centra (eight percent). The caudal centra were out of line in only nine percent of the skeletons, corroborating the observation that the tails usually appear normal at birth. The vertebrae were not severely affected and seldom were more than two adjacent vertebrae involved in fusions.

The ribs were the most consistently affected part of the skeleton (Figure 9). Fusions were common, usually somewhat distal to the vertebrae, infrequently involving them even when the ribs were extensively fused. One hundred eighteen out of the 139 abnormal skeletons examined, or 85 percent, had unilateral or bilateral rib fusions. The two sides were equally affected, while the incidence of animals with bilateral fusions was approximately twice that of those having unilateral effects. Several other rib anomalies were also found in *Rfrf* skeletons. These included reduction in the length or width of the ossified portion of a rib, crowding, occasional absence of a rib, and, in one case, the lack of a costal cartilage for an otherwise normal rib. In five animals, knobs

TABLE I. Genetic ratios obtained by classification on the basis of the presence of fused ribs

Parental phenotypes ♀ × ♂	No. of offspring	No. having fused ribs	% fused ribs	χ^2
Kinky × Kinky	183	126	69	(3:1)3.688, (2:1)0.393
Kinky × Normal	229	92	40	(1:1)5.000
Normal × Kinky	379	132	35	(1:1)34.
Normal × Normal	53	9	2	



SKELETONS OF NORMAL AND MUTANT RIB FUSIONS

Figure 9

These drawings of alizarin-stained skeletons of newborn animals illustrate (left to right) a normal mouse (*rfrf*), a heterozygote (*Rfrf*), and a presumed homozygote (*RfRf*). The *Rfrf* skeleton has fused ribs but otherwise is nearly normal. In the *RfRf* skeleton, note the extensive fusion of ribs and neural arches, fragmentation of centra, reduction in size, and termination of the vertebral column in the sacral region.

giving the appearance of extra rib heads were found opposite the capitular heads. Cervical ribs were encountered so frequently (74 percent) in both *Rfrf* and *rfrf* animals as to constitute a normal deviant, not an effect of the gene *Rf*. Ninety-six percent of all *Rfrf* and 99 percent of the *rfrf* skeletons had the normal number of 13 pairs of thoracic ribs. The sternebrae were normal in all *Rfrf* skeletons examined.

Abnormalities of the appendicular skeleton and of the bones of the skull were not observed in any *Rfrf* skeleton. The first sacral vertebra is not distinguishable at this age; therefore the serial

TABLE II. Sixty-day weights of *Rfrf* and normal-tailed mice, from the same litters

Phenotype	Sex	No. animals	Weight range	Average weight
Rfrf	♂♂	22	15-23 gms.	18.0 gms.
	♀♀	35	13-19	15.3
Normal	♂♂	19	16-24	20.2
	♀♀	19	12-21	17.1

number of the first vertebra to be intersected by a line through the anterior borders of the ilia was tabulated for all skeletons. This line intersected the twenty-sixth vertebra in 94 percent of all *Rfrf* and *rfrf* skeletons examined.

Five extremely abnormal animals, from mating of *Rfrf* × *Rfrf*, were found dead shortly after birth. They had short, wide bodies; three were tailless and two had rudimentary, filamentous tails. Two were also exencephalic. The skeletons revealed more extensive anomalies of the axial skeleton than had been seen previously (Figure 9). The vertebral components were so jumbled as to make alignment almost impossible. The centra were fractured, and often pieces of several vertebrae were fused together, especially in the thoracic and lumbar regions. Fusions of adjacent neural arches occurred along the whole length of the vertebral column, often involving a large number of vertebrae. The vertebral column stopped in the mid-sacral region, without any evidence of caudal vertebrae, in all of the skeletons. Spina bifida of the lower part of the body occurred in one animal. In the exencephalic animals, the lateral occipitals were fused with the neural arches of the first cervical vertebra. The thoracic basket was short and wide, with extensive bilateral rib fusions, and apparent reduction of the number of ribs. Several or all of the sternebrae were also fused. An additional exencephalic animal was found in one litter containing these very abnormal animals. Its skeleton was normal, except for a pair of fused ribs, and it was classified as *Rfrf*. The occurrence of exencephaly in the heterozygote might account for the deficiencies of *Rfrf* (Table I).

These five extremely abnormal animals, which occurred in four litters of 22 young, represent the homozygous class, *RfRf*.

Older animals

Forty-five abnormal skeletons, ranging in age from five days to 18 months, of strain 129 and of its hybrids with C3HeB/FeJ and BALB/cJ, were also studied. The types and frequencies of abnormalities were similar to those described for newborns and term embryos, were also limited to vertebral column and ribs, and included: fusions of adjacent neural arches (67 percent), fusions of adjacent centra (two percent), hemicentra (two percent), bilateral bodies in the centra separate from each other (two percent) or joined by a bony isthmus (four percent), and thoracic curvatures and displaced centra (36 percent). In addition, there were several other anomalies, which become apparent with age: incomplete closure of neural arches (18 percent), reduction or absence of the spine on the ninth vertebra (nine percent), extra thoracic and cervical arch pieces (two percent), and asymmetrical caudal transverse processes (two percent).

Thirty-one (69 percent) of the *Rfrf* skeletons had both fused ribs and kinky tails, 10 (22 percent) had fused ribs and normal tails and four (nine percent) had normal ribs and kinky tails. Fused ribs, not tail kinking, is the more regular manifestation of the gene.

Breeding Performance

Breeding records were kept on 76 kinky- and 52 normal-tailed females of the 129 *Rf* colony. The incidence of sterility in kinky-tailed females is twice as great (55 percent) as it is for normal-tailed females (25 percent). When autopsied at four to six months of age, sterile animals had uteri which showed little evidence of hormonal stimulation. There were no differences between kinky- and normal-tailed fertile females in reproductive capacities. The average number of litters produced in a female's lifetime was four, and the average litter size was five, for both phenotypes. Normal females had their first litters by the time they were three and one-half months of age, kinky-tailed females by four months.

Discussion

Evidence has been presented for a new semidominant mutation which causes fused ribs and/or kinky tails. When two affected animals are mated, 69 percent of their off-

spring are similarly affected (Table I), which indicates lethality of the homozygote. Further evidence for lethal action of *RfRf* is provided by the fact that none of the 32 kinky-tailed animals tested were found to be genetically homozygous. Additional evidence for lethality of *RfRf* is provided by the five very abnormal animals, offspring of two heterozygotes, which are considered to be homozygotes, since the skeletons are more extremely affected than those of heterozygotes.

A number of other mutations have been described for the mouse which result in tail kinking, some of which also produce fused ribs. Homozygous tail-kinks (*tktk*) have shortened, kinky tails, and fusions and fragmentations of the cervical and upper thoracic vertebrae, with little involvement of those between[2]. The first two pairs of ribs and the first two sternebrae are anomalous as a result. The pelvic girdle is shifted one vertebra posteriorly in *tktk*. The *Rf* gene, at least in the heterozygous state, exerts no effect on the number of presacral vertebrae. Stub (*sbsb*), apparently viable when homozygous, has a shortened tail and a short, wide thoracic basket with reduction in the number of ribs and fusions between them[1]. Diminutive (*dmdm*) also exerts an effect upon the ribs, regularly adding a pair articulating with the twenty-first vertebra[5]. Fused ribs occur less frequently in *dmdm* than they do in *Rfrf*, while kinky tails are a constant manifestation of the *dm* syndrome. Loop-tail homozygotes (*LpLp*) have extensive neural arch fusions, but the centra are normal except in the thoracic region, where a pronounced twist of the body is believed to be responsible for the fragmentation of the thoracic centra, and for the fusions and reduction in the number of ribs[4]. The sternebrae are fused into a single bone, because of the inability of the ribs to make contact with the developing sternal bands.

The effects of the gene Rib fusions in the heterozygous state are primarily limited to the tail and ribs, with the majority of *Rfrf* animals having both fused ribs and kinky tails. Heterozygotes are slightly smaller at 60 days than normals (Table II), but there is not as pronounced a size difference as is found in diminutive mice[5].

The *Rfrf* animals, regardless of their genetic background, have similar characteristics, unlike Tail-short (*TsTs*), which exhibits marked differences in the expressivity of the gene on various backgrounds[3]. Affected animals have short tails in the strain of origin, but outcross offspring were tailless on one background, normal-tailed on another, and both short-tailed and tailless in varying numbers on still others. Morgan[3] believed modifiers were present within the inbred strains he used, which could vary the phenotypic expression of the Tail-short gene.

The gene *Rf* has pleiotropic effects on the formation of the somites and the development of the central nervous system[6]. Homozygotes may be recognized at eight to nine days of gestation when they are retarded in development, lack somites, and have irregular waves in the neural plate and tube. The extensive rib fusions in homozygotes are traceable to the failure of somite formation. Heterozygotes may be recognized at 12 days of gestation when sporadic fusions of rib anlagen may be seen in frontal sections. These fusions are probably the result of abnormalities of the ventro-lateral extensions of otherwise normal somites.

Summary

The effects of a new mutation in the mouse, named Rib fusions and symbolized *Rf*, have been described. *Rf* is a semidominant autosomal gene which is lethal when homozygous. Heterozygotes are characterized by fused ribs and/or kinky tails. The few animals classified as homozygotes have a shortened vertebral column, and a short, wide thoracic basket with extensive neural arch and rib fusions.

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CYTOLOGICAL INVESTIGATIONS IN THE GENUS BROMUS

III. The Cytotaxonomic Significance of the Satellite Chromosomes

JURGEN SCHULZ-SCHAEFFER*

SATELLITE chromosomes are the most conspicuous type of chromosome, and they are of value for cytological comparisons.

Tischler[22] states that the genomes of all higher plants contain at least one satellite chromosome. In some species the number of satellite chromosomes appears to be less than the number of "basic genomes".† There are various reasons for this discrepancy. In some plants satellites may be present but not discernible by present techniques, or satellites may be attached so closely to the main chromosome body that only extreme pressure on the cover glass will separate them from the remainder of the chromosome. This latter is particularly true of the larger satellites. Gates[3] has reported that with increasing size the satellites are more closely connected to their chromosomes. The author found that in *Secale montanum* the large tandem satellite of chromosome 4 is frequently not discernible. It is so closely attached to chromosome 4 and the secondary and tertiary constrictions are so obscure that the short arm and the tandem satellite together often appear as one long arm[10]. Another reason for the lack of satellite chromosomes could be the occurrence of chromosome mutations or chromosome substitutions during the course of evolution. This also could explain the occurrence of a higher number of satellite chromosomes than expected.

The secondary constriction of a satellite chromosome, also called the nucleolar organizer region, seems to function specifically in connection with the transfer of material to and from the chromosomes during the nuclear cycle. Accordingly, it seems that every nucleus must have at least one such chromosome in order to pass through a normal cycle[16]. Consequently, all diploid ancestors of our auto- and alloploid species must have once possessed at least one pair of satellite chromosomes. Theoretically, tetraploids should have at least two, hexaploids at least three, and octoploids at least four pairs of homologous satellite chromosomes, and so on. Structural and physiological changes may have occurred during evolution. Chromosomes originally able to organize nucleoli may have lost that ability. The special function of organizing nucleoli may then have been borne by fewer satellite chromosomes. In the present study of 31 *Bromus* species, the number of satellite chromosomes (160) almost coincides with the number of basic genomes (166), confirming the author's belief that this is a functionally significant pattern suggestive of an evolutionary "original" pattern.

If it is assumed that there is a characteristic satellite chromosome representing each basic genome, this satellite chromosome can serve as an *indicator chromosome* and in this way be a valuable asset in cytotaxonomic comparisons. Such satellite chromosomes usually are

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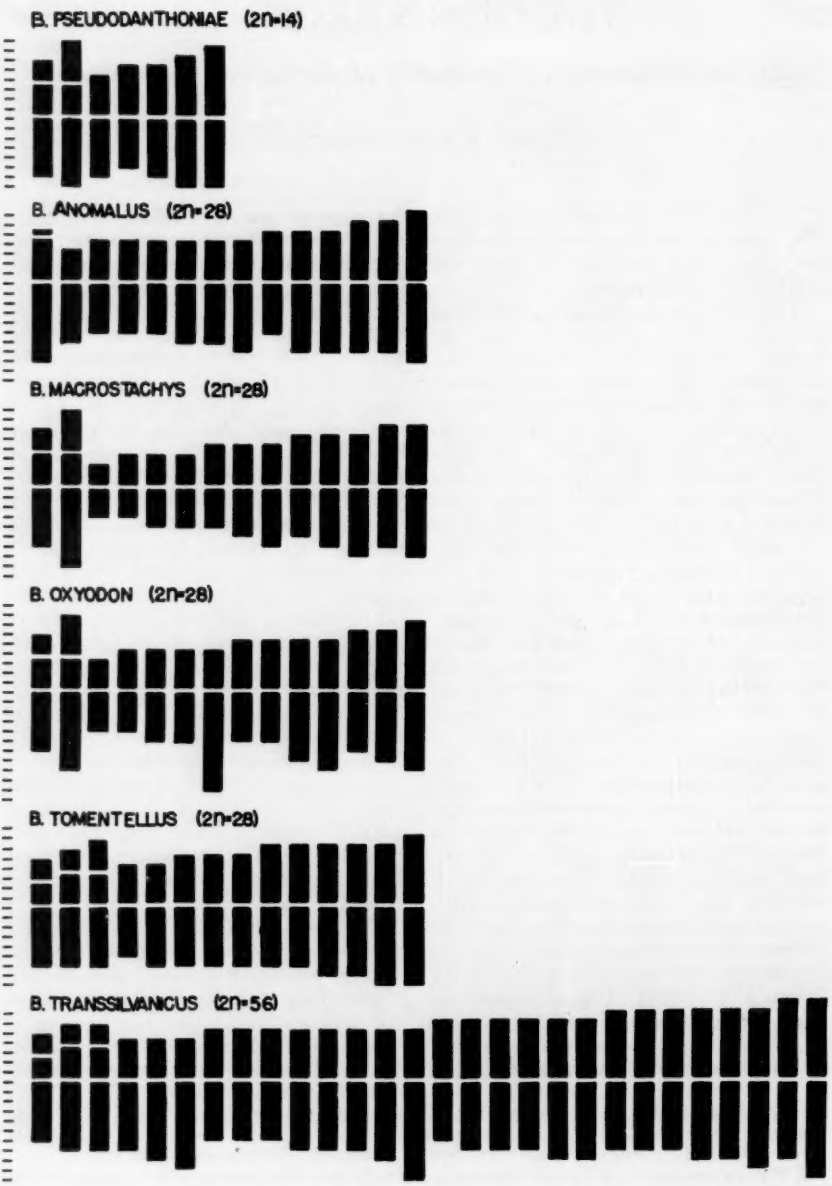
†"Basic genomes"—groups of chromosomes which have been present in the gametes of the diploid ancestors of polyploids and those groups which are present in the gametes of the still existing diploids of a genus.


B. PSEUDODANTHONIAE (2n=14)
B. ANOMALUS (2n=28)
B. MACROSTACHYS (2n=28)
B. OXYODON (2n=28)
B. TOMENTELLUS (2n=28)
B. TRANSSILVANICUS (2n=56)

characterized sufficiently by unique structure, so that their homology can be definitely established in the genomes of different species. The present study was designed to determine whether such relationships exist in the genus *Bromus*.

Materials and Methods

Seeds of the *Bromus* species investigated cytologically were obtained from plant material centers and from various botanic gardens. A compilation of all *Bromus* species discussed in this paper is given in Table I. The species are arranged alphabetically. Accession numbers and names of seed distributors, as well as locations where the species were collected, are reported. Chromosome numbers are listed in accordance with observations made during the course of the study.

Root tips were examined from potted plants and from seeds germinated in petri dishes. The pretreatment was made according to the oxyquinoline technique of Tjio and Levan[23], and as modified by Markarian and Schulz-Schaeffer[9]. The cold treatment method of Delaunay[1] was also used.

Measurements of root-tip chromosomes were made with an eyepiece micrometer. The measurements represent averages from a minimum of 10 chromosomes in each species.

Results and Discussion

Chromosome numbers of 42 taxa within the genus *Bromus* were reported previously[12, 14]. In six of the 31 taxa compared in the present study chromosome numbers were not reported previously. These are the species *B. pseudodanthoniae* ($2n=14$), *B. anomalus* ($2n=28$), *B. macrostachys* ($2n=28$), *B. oxyodon* ($2n=28$), *B. tomentellus* ($2n=28$), and *B. transsilvanicus* ($2n=56$). Karyotypes of these six species are presented in Figure 10. Photomicrographs of *B. pseudodanthoniae*, *B. anomalus*, and *B. macrostachys* are presented in Figures 11 and 12.

The 31 taxa which were compared in this study on the basis of the shape of their satellite chromosomes belong to the sections *Bromopsis*, *Bromium*, and *Ceratochloa*. Fourteen types of satellite chromosomes, found in an earlier satellite chromosome comparison of 20 *Bromus* species[14], have been designated by Roman numerals I to XIV. Type VIII has since been omitted because it could not be substantiated through further studies. Two new types, IIa and VIIa, were found in this study, so that the total number of satellite chromosome types found in *Bromus* species is now 15. In Figure 13 drawings of IIa, VI, VIIa, and X, and photographs of the other 11 are arranged according to the lengths of the satellites. The lengths of the satellites vary from 0.25μ in I to 2.5μ in XII, XIII, and XIV.

Of particular significance is the shape of the satellite chromosomes II, VI, VII, IX, X, XII, and XIV in *Ceratochloa*. Their satellited short arms are all extremely short (0.5 to 1μ), which distinguishes them from the satellite chromosomes in the other sections. The only type with a similar short arm is type IIa of *Bromopsis*, which is represented in *B. transsilvanicus* and *B. tomentellus*. This cytological differentiation of the members of *Ceratochloa* is in conformity with morphological observations. *Ceratochloa* is characterized in having species with three-lobed ovaries and frequent occurrence of three stigmata[17]. The spikelets are distinctly oppressed laterally. These are characteristics not common to any other section of the genus.

Typical satellite chromosome types were also found for each of the other two sections investigated in this genus. The types I, IIa, III, and IV in *Bromopsis* are smaller satellite chromosome types with very short satellites. The larger satellite chromosome types (V and XI), and particularly the type with a long satellite (XI), were found to predominate in *Bromium*. These intrinsic cytological differences are associated with a differentiation in morphological characteristics. The species within *Bromopsis* are all perennials and are different in this respect from those in *Bromium* which are exclusively annuals. Satellite chromosome types VIIa and XIII have been found only in one species each. Va and XIa

IDIOGRAMS OF SIX *BROMUS* SPECIES

Figure 10

Each column represents two homologous chromosomes. The satellite chromosomes are placed at the beginning of the idiograms and are arranged according to the length of their satellites. The rest of the chromosomes are arranged according to the length of their short arms. One unit on the scale left of the idiograms equals 0.4 mikrons.

TABLE I. Sources of collections and chromosome numbers in *Bromus*

Species	Acc. No.*	Source	2*n* (own observation)
B. aleutensis Trin. et Griseb.	B-1	Botanic Gardens of the University of Copenhagen, Denmark.	56
B. anomalus Rupr.	P.I. 232,200	Yellowstone National Park, Wyoming.	28
B. arvensis L.	Gr. 224/52	Botanic Gardens of the University of Marburg-Lahn, Germany. Identified by J. Schulz-Schaeffer in 1956.	14
B. brizaeformis Fisch. et Mey.	J-19	Blue Mountains, east of Milton, Oregon. Collected on July 8, 1954, by J. P. Meiners. Identified by J. Schulz-Schaeffer in 1956.	14
B. catharticus Vahl.	B-5	U.S.D.A., Yuma Valley and Mesa Experiment Stations, Arizona.	42
B. ciliatus L.	B-6	Botanic Gardens of the University of Copenhagen, Denmark.	14
B. coloratus Steud.	B-7	Royal Botanic Gardens, Kew, England.	42
B. commutatus Schrad.	P.I. 228,399	Kuhe Sion between Saral and Ardebil Azerbaijan, Iran, 1955. Collected by H. S. Gentry. Identified by J. R. Swallen in 1957.	14
B. danthoniae Trin.	P.I. 141,382	Cheshmashirin, Bakhtiari, Iran.	14
B. erectus Huds.	Gr. 29/51	Botanic Gardens of Helsinki, Finland. Identified by J. Schulz-Schaeffer, in 1956.	56
B. haenkeanus (Presl.) Kunth	P.I. 197,850	Estacion Experimental Pergamino, Pergamino, Argentina (orig. from Mendoza).	42
B. inermis Leyss.	Gr. 267/51	Eugen Vogt (Schlusselburg), DSG, Springe/Hannover, Germany. Identified by J. Schulz-Schaeffer in 1956.	56
B. intermedius Guss.	P.I. 107,506	(Introduced as *B. severtzowi* Regel in 1934) Westover-Enlow expedition, Bot. Gard., Taskent, Turkistan.	28
B. japonicus Thumb.	B-14	Botanic Gardens of the University of Marburg-Lahn, Germany. Identified by J. Schulz-Schaeffer in 1956.	14
B. laciniatus Beal.	B-15	Royal Botanic Gardens, Kew, England.	56
B. lepidus Holmb.	B-16	Royal Botanic Gardens, Kew, England.	28
B. macrostachys (L.) Desf.	P.I. 203,451	Valley near Ahiboz, 25 miles south of Ankara, Turkey.	28
B. maritimus (Piper) Hitchc.	B-18	Mr. Beecher Crampton, Davis, California. Collected summer of 1954 on the coastal bluffs at San Francisco, California.	56
B. oxyodon Schrenk	P.I. 220,377	Road between Baghlan and Kundus, Kataghan, Afghanistan, 1954. Identified in Dec. 1955, by J. R. Swallen, U. S. Nat. Herbarium.	28
B. polyanthus Scribn.	P-7477	Albuquerque, New Mexico, 1937.	56
B. pseudodanthoniae Drobov.	P.I. 211,006	Fifty miles west of Kunduz, Kataghan Province, Afghanistan.	14
B. riparius Rehm	P.I. 98,277	(Introduced to the U. S. under the name *B. erectus* Huds. in 1932) Samara, European Russia. Identified by J. R. Swallen, U. S. National Herbarium, April, 1960.	70
B. riparius Rehm × *B. inermis* Leyss.	Gr. 125/53	(Introduced to Germany as *B. erectus* Huds.) Botanic Gardens, Sacavem, Portugal. Identified by F. C. Elliott, in 1956.	63
B. secalinus L.	B-24	Botanic Gardens of the University of Copenhagen, Denmark.	28

have been reported previously[13] but could not be identified in more recent studies.

In retrospect there appear to be satellite chromosome types representing genomes which are *specific* for each of the three sections investigated. These are types III in *Bromopsis* (Figure 14), XI in *Bromium* (Figure 14), and IX in *Ceratochloa* (Figure 14). Type III is present in six of eight *Bromopsis* species, type XI in nine of 12 *Bromium* species, and type IX in eight of 11 *Ceratochloa* species. Roman numerals representing the satellite chromosome types marking *specific* genomes for each section are encircled in Figure 14.

Figure 14 is an attempt to show satellite chromosome similarities within the genomes of the *Bromus* species investigated. The ploidy level is indicated by the number of circles around the species names. Diploid species are represented by one circle, tetraploid species by two circles, and so on. Species groups with similar patterns are arranged on one horizontal line of the diagram. Straight lines connecting circles indicate satellite chromosome similarities. In general, the satellite chromosome type marking the similarity of a group is the first or last one listed in a circle. For instance, in section *Bromium*, type III is characteristic of the species *B. macrostachys, B. commutatus, B. secalinus, B. oxyodon,* and *B. pseudodanthoniae,* which distinguishes this group from the one below. The second group has satellite chromosome type V in common and contains *B. japonicus, B. intermedius, B. squarrosus,* and *B. danthoniae.* Both groups mentioned have satellite chromosome type XI in common.

Besides being *specific* for *Bromopsis,* satellite chromosome type III was also found in five species of *Bromium.* These species, *B. macrostachys, B. commutatus, B. secalinus, B. oxyodon,* and *B. pseudodanthoniae,* conceivably may be amphiploid sectional hybrids between *Bromopsis* and *Bromium* ancestors. Hitchcock and Chase[6] state that the species of this group, containing *B. secalinus, B. commutatus, B. mollis,* and *B. racemosus,* are closely allied, differentiated only by arbitrary characters. The forms are recognized as species in most recent European floras, and Hitchcock and Chase followed this disposition. Here the cytological observations of satellite chromo-

TABLE I. Sources of collections and chromosome numbers in *Bromus* (Continued)

Species	Acc. No.*	Source	2*n* (own observation)
B. sitchensis Trin.	B-25	Botanical Gardens, Berlin-Dahlem, Germany.	42
B. sitchensis Trin.	B-26	Botanical Gardens of the University of Rostock, Germany.	56
B. squarrosus L.†	F-276	Mr. Jack P. Meiners, U.S.D.A. Regional Smut Laboratory, Dept. of Plant Pathology, Washington State University, Pullman, Washington, 1956.	14
B. tomentellus Boiss.	P.I. 111,530	Institute of Plant Industry, Leningrad, U.S.S.R. Collected by Westover-Enlow expedition in 1934.	28
B. transsilvanicus Schur.	Gr. 184/53	Agricultural Botanic Gardens, Budapest, Hungary.	56
B. valdivianus R.A. Phil	P.I. 197,852	Estacion Experimental Pergamino, Pergamino, Argentina (originally from Nahuel Huapi).	42
B. viviparus Rehm.	B-31	Botanic Gardens of the Friedrich Schiller University, Jena, Germany.	56

*Explanation of accession numbers.

B = Seed available from the Department of Agronomy and Soils, Montana State College, Bozeman, Montana.

P.I. = Seed available from U.S.D.A., A.R.S., New Crops Research Branch, Crops Research Division, Plant Introduction Stations, Western Region, Pullman, Washington, and North Central Region, Ames, Iowa[11].

Gr = Seed available from Institut fuer Kulturpflanzenforschung, Abteilung Systematik und Sortiment, Gatersleben, Kreis Aschersleben, Germany.

J,F = Seed available from U.S.D.A., Regional Smut Research Laboratory, Division of Plant Pathology, Washington State University, Pullman, Washington.

P = Seed available from U.S.D.A. Soil Conservation Service, Pacific Region, Plant Material Center, Washington State University, Pullman, Washington.

† P.I. 108,469. Collected by H. L. Westover and C. R. Enlow in the Ukraine, Russia, 1934.

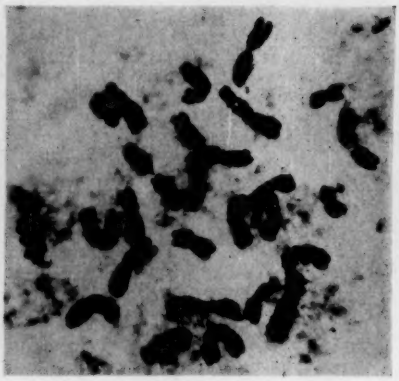
***BROMUS* CHROMOSOMES**

Figure 11

A—B. pseudodanthoniae (2*n*=14). *B—B. anomalus* (2*n*=28). 1200×.

somes agree with the morphological studies.

Octoploid *B. sitchensis* (*Ceratochloa*) is interpreted to be an amphiploid sectional hybrid between *Ceratochloa* and *Bromium* ancestors (Figure 14). It possesses satellite chromosome type XI, which is *specific* for *Bromium*, substantiating such a hypothesis. Autoalloploid character is postulated to be expressed in *B. transsilvanicus* (IIa, III, III), *B. erectus* (III, III, III, and IV), *B. riparius* (I, III, III, and IV), and *B. inermis* (I, III, and III) with the main contributing genome being the one carrying type III. Stebbins[18] (p. 315) explains the theoretical



B. MACROSTACHYS

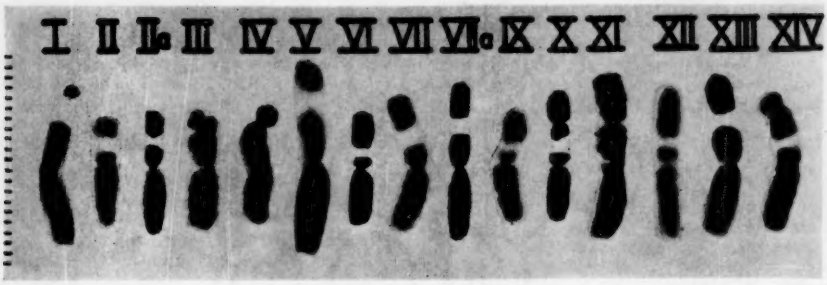
Figure 12

Photomicrograph at 1280×.

formation of autoallooctoploids. The possible origin of *B. riparius* (I, III, III, and IV) from a cross between ancestors of *B. inermis* (I, III, and III) and *B. ciliatus* (IV) was discussed previously[14].

Changes in the appearance of metaphase chromosomes such as the satellite chromosomes compared in this particular study may be brought about chiefly by chromosome mutations, such as deficiencies, duplications, translocations, and inversions. These structural alterations could result in parallel changes or even convergent resemblances of satellite chromosomes in unrelated lines. The data presented support the hypothesis that such coincidental changes have not occurred to a remarkable extent for the satellite chromosomes of the genus *Bromus*. The distribution of satellite chromosome types is in conformity with major morphological characters expressed, for instance, in the sectioning of the genus. As systematicists have used morphological plant characters almost exclusively as the criterion for their studies in the past, the now existing sections within the genus are an expression of morphological separation only.

The method of determining interrelationships among basic genomes of a genus, by using satellite chromosomes as indicator chromosomes, would appear to be a promising one. Further investigation of the method is needed. It could conceivably contribute to a more complete evaluation of evolutionary interrelationships among species. It could supplement the method of determining intrageneric relationships by interspecific hybridization with subsequent analysis of meiotic pairing, which has been described by Kihara and Nishiyama[7] and was then called "Analysatoren Methode."



SATELLITE CHROMOSOMES

Figure 13

Arranged according to the length of the satellites are satellite chromosome types I to XIV of 30 species and one interspecific hybrid of *Bromus*. Types IIa, VI, VIIa, and X are presented as drawings. One scale unit equals 0.4 mikrons.

This method has been used by Stebbins and co-workers[19, 20, 21] for genome analysis in the genus *Bromus*. The method of interspecific hybridization takes a great amount of time and effort and sometimes hybrids cannot be obtained between certain species. Furthermore, complete association of all chromosomes able to pair is expected to take place only with the favorable occurrence of unusual genic and environmental or physiological conditions, which occur very rarely[4]. Sears[15] warned of a too inflexible application of the classical genome analytical method based on pairing observations. He states that this method is open to some criticism because of the possibility of nonhomologous association, such as found by Levan[8] in haploid rye.

Elliott[2] found normal pairing between three basic genomes (21 bivalents) in a hybrid between *B. inermis* ($2n=56$) and *B. riparius* ($2n=70$) while the rest of the chromosomes were represented as uni- and multivalents. This seems to support the method of using indicator chromosomes, since *B. inermis* and *B. riparius* have three satellite chromosomes in common (types I, III, and III) representing three basic genomes. More interspecific hybridization with subsequent genome analysis within the genus *Bromus* could furnish evidence for evaluating the applicability of the indicator chromosome theory. However, the most complete picture of species interrelationships will always be the one which has been derived by using all the different approaches known at a certain time. To date, we can use the morphological, serological, and cytological approaches such as chromosome pairing and chromosome morphology. Other methods of approach may be discovered in the future which may give an even more complete picture of species interrelationships. One of those newer approaches in using the serological method is the immunological method as proposed by Gell *et al.*[5]. It divides the investigated species into well-defined groups in a similar manner to the "indicator chromosome" method. If the relationships between the species of *Bromus* indicated here are substantiated, the indicator satellite chromosome method of using satellite chromosomes to represent basic genomes might be extended for similar analysis of other taxonomic categories.

Summary

Thirty *Bromus* species and one interspecific hybrid belonging to the sections *Bromopsis*, *Bromium*, and *Ceratochloa* were compared cytologically. The criterion for comparison was the morphology and size of the satellite chromosomes. Fifteen types of satellite chromosomes were found and are discussed.

A relationship of the results of the satellite chromosome method of analysis to taxonomic categories is postulated.

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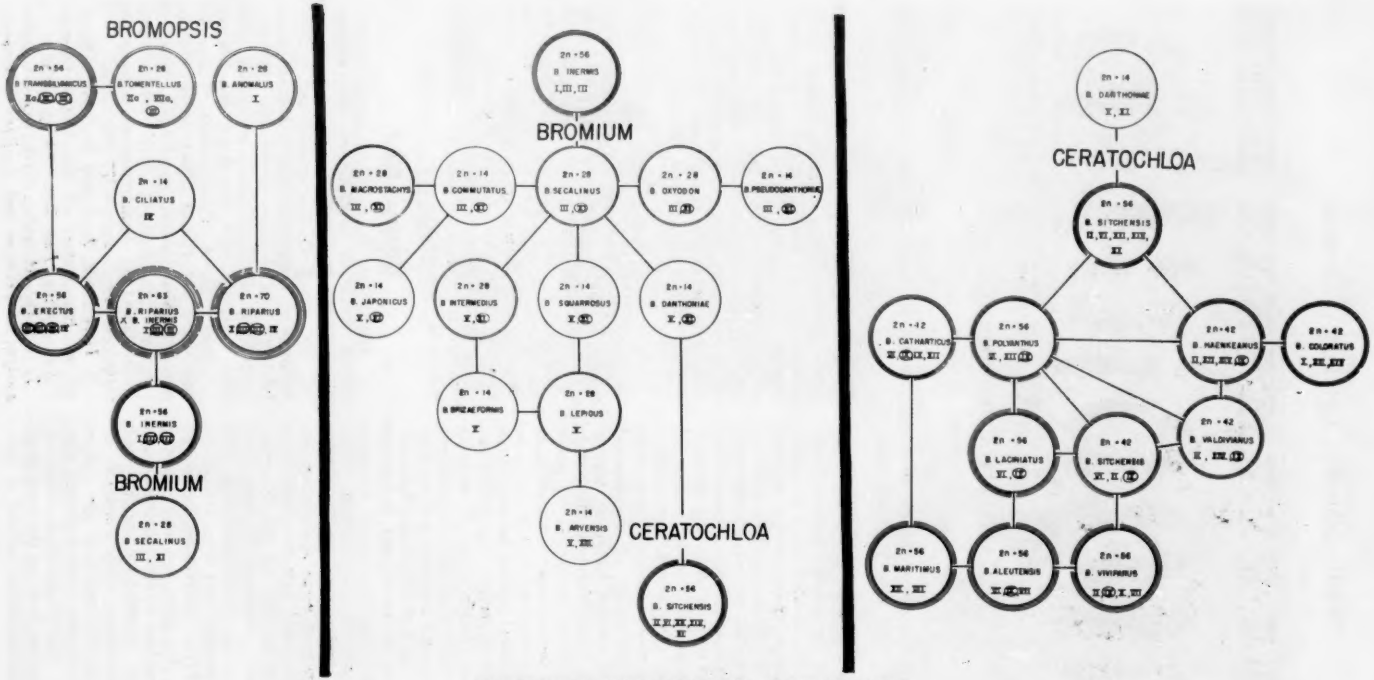

SATELLITE CHROMOSOME SIMILARITIES

Figure 14

Concluded from the satellite chromosome analysis is this diagrammatic representation of satellite chromosome similarities among 30 species and one interspecific hybrid of *Bromus*. The ploidy level is indicated by the number of circles around the species name. Roman numerals indicate satellite chromosome types. Straight lines connecting circles indicate satellite chromosome similarities. Represented are (left) seven species and one interspecific hybrid of section *Bromopsis*, (center) 12 species of section *Bromium*, and (right) 11 species of section *Ceratochloa*.

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Correction—Interspecific Hybrid in *Paspalum*

Two errors appeared in the recent paper by Hugh W. Bennett and E. C. Bashaw, "An Interspecific Hybrid in *Paspalum*" (*Jour. Hered.* 51:81-85). On page 81, the sixth sentence of the first paragraph in column two should read, "The pollen plants were removed from the control room and quickly placed next to the plants to be pollinated and their flowering racemes placed in contact with the emasculated racemes." On page 84, the first sentence of the first paragraph in column two should read, "The range of chromosome numbers found in the F_2 progeny and the variation in the appearance of the plants indicate that the F_1 reproduced sexually."

The editor regrets these errors.

INFLUENCE OF THE DWARF GENE ON INSULIN SENSITIVITY

of Beef Cattle

C. W. Foley, C. J. Heidenreich and J. F. Lasley*

SEVERAL kinds of hereditary dwarfism have been reported in beef cattle, but the one known as "snorter" dwarfism is of the greatest economic importance. It is inherited as a simple Mendelian recessive trait[10, 12, 16].

Efforts to determine the actual physiological cause of "snorter" dwarfism have not been too successful. It has been reported that dwarfs are deficient in the thyrotropic hormone[2], but studies in which radioactive iodine was used as a tracer led to the conclusion that the secretion rates of thyroxine by the thyroid and of the thyroid stimulating hormone by the anterior pituitary gland were within the normal range[3]. Other studies[13] indicate that the pituitaries of dwarf cattle contain normal amounts of ACTH and the growth hormone. Histological studies, however, show that some dwarfs possess cystic pituitaries and adrenals, which suggests that these glands may often be abnormal in the dwarf animal[7].

Preliminary studies at the Missouri Agricultural Experiment Station suggested that "snorter" dwarfs possess an inborn error in carbohydrate metabolism[8, 14]. To investigate this further an insulin sensitivity test was conducted with animals of the three different genotypes to obtain a clearer understanding of the abnormal physiological mechanism which is possibly involved.

Materials and Methods

Data were obtained from 36 animals. This number included 12 mature pedigree-clean cows, 11 mature cows and one bull (which were known to be carriers of the dwarf gene) and 12 dwarfs. Nine of the 12 dwarfs were considered mature. Two of the dwarfs were Angus and 10 were Herefords, and they were equally divided in number between the two sexes. The dwarfs and carriers were kept at the same location and the pedigree-clean cows were kept at another. Because of the time and labor involved, blood collections were made from the individual animals over a period of several weeks.

Regular protamine-zinc insulin was injected into the external jugular vein of the animals at a dosage level of 0.8 unit of insulin per kilogram of body weight. Prior to, and following the injection of insulin, approximately 10 milliliters of blood were obtained from the animals. Potassium oxalate tablets were used as an anticoagulant.

The initial blood sample obtained prior to the administration of insulin was considered as a control. The additional blood samples were collected at the following time intervals: 30 minutes, 1, 2, 6, 10 and 12 hours subsequent to the injection of insulin. Forty-eight hours after the initial injection of insulin, the exact procedure, with one exception, was again followed on the same animals. At this time, 0.3 unit of insulin per kilogram of body weight was used instead of 0.8 unit. The blood sugar response at the different dosage

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levels was compared for the three genotypes and was used as a measure of insulin sensitivity.

Blood glucose determinations were made by both the Nelson-Somogyi and Folin-Wu methods in each of the 504 blood samples collected from the 36 animals used in this study. These replicate determinations by two different methods were made on each sample in order to double check the results and to compare the glucose values obtained by the two different methods of blood sugar analysis.

Results and Discussion

Average blood glucose values for blood samples determined by the two different methods in dwarf, carrier and pedigree-clean animals are shown in Tables I and II. The statistical analysis of the data is summarized in Table III.

A highly significant difference in blood sugar values was found to exist between the two methods of analysis used in this study. For the pedigree-clean and the carrier animals the Folin-Wu method usually gave higher blood sugar values than did the Nelson-Somogyi method. This difference observed between the two methods appeared to be greater for the pedigree-clean animals than for the carriers of the dwarf gene and was consistent at the two levels of insulin administered. Little difference was noted between the two methods for dwarfed individuals. These results suggest that a reducing compound was present in the blood of normal appearing animals that was lacking in the blood of the dwarfs. This deficient substance would be one or more of several substances, including creatinine, uric acid, thioneine, ergothioneine, or glutathione which most often account for the increased values obtained with the Folin-Wu method (tungstic filtrate) over those obtained with the Nelson-Somogyi method (zinc filtrate)[1, 11, 18].

TABLE I. Mean blood sugar values in mg. percent as determined by the Folin-Wu and Nelson-Somogyi methods of analysis (.8 unit of insulin per kilogram body weight)

Time after insulin	Clean		Carrier		Dwarf	
	N.S.	F.W.	N.S.	F.W.	N.S.	F.W.
Initial	60.9	80.2	54.0	61.7	51.6	50.8
½ Hour	34.0	51.8	32.0	35.5	23.2	18.7
1 Hour	30.2	50.0	31.5	30.7	27.3	19.8
2 Hours	27.1	44.7	29.9	35.4	28.4	24.8
6 Hours	42.2	53.4	33.9	37.1	28.7	26.2
10 Hours	61.3	75.6	42.5	46.6	39.2	37.4
12 Hours	67.4	86.8	49.3	55.0	48.6	43.2
Averages	46.16	63.21	39.01	43.14	35.29	31.55

Differences in blood sugar values and blood sugar changes following insulin administration also were found to exist among cattle of the three genotypes. When 0.8 unit of insulin per kilogram of body weight was administered and the blood sugar determinations made by the Nelson-Somogyi method of analysis, it was observed that a highly significant difference ($P<.01$) existed among the means for all three genotypes. At the lower dosage level of 0.3 unit of insulin per kilogram of body weight, a highly significant difference ($P<.01$) existed between the dwarfs and the two other genotypes concerned, but there was no significant difference between the carriers and the pedigree-clean animals. Similar results were also obtained with the Folin-Wu method at both dosage levels except that there was a significant difference ($P<.05$) between carrier and pedigree-clean animals at the lower level of insulin administration.

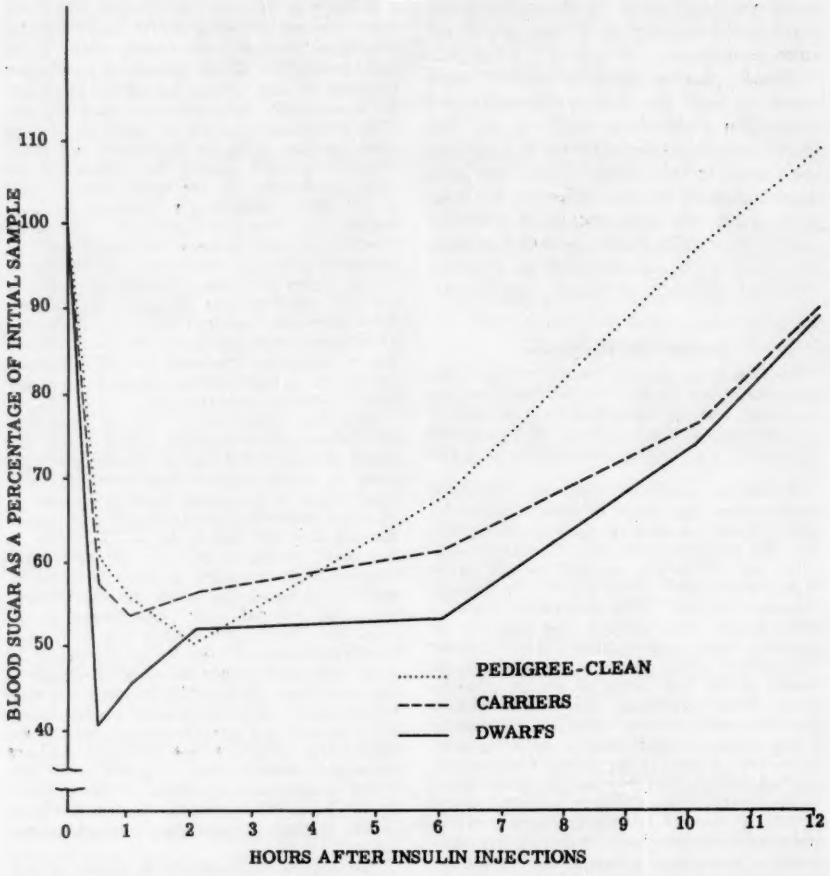
Data also presented in Table I show that the pedigree-clean animals, when given the larger dosage of 0.8 unit of insulin per kilogram of body weight, had returned their blood sugar to the initial level at the end of 12 hours, whereas both the dwarfs and carrier animals were not able to do so. However, at the lower dosage of 0.3 unit of insulin per kilogram of body weight, as shown in Table II, both the pedigree-clean and the carrier animals had restored their blood sugar to the initial level by the end of eight hours, whereas the dwarfs had not done so by the end of 12 hours. This is demonstrated more effectively in Figures 15 and 16 in which the data for each blood sample following insulin administration are expressed as a percentage of the initial blood sugar level. The fact that the three different genotypes varied in their response to the different dosage levels of insulin is further verified in Table III in which a highly significant insulin dosage × genotype interaction was found ($P<.01$).

The degree of sensitivity to insulin in animals has been measured in several different

TABLE II. Mean blood sugar values in mg. percent as determined by the Folin-Wu and Nelson-Somogyi methods of analysis (.3 unit of insulin per kilogram of body weight)

Time after insulin*	Clean		Carrier		Dwarf	
	N.S.	F.W.	N.S.	F.W.	N.S.	F.W.
48 Hours	59.0	76.5	59.0	67.6	47.8	50.8
48½ Hours	30.7	52.1	29.5	35.0	24.8	22.4
49 Hours	31.1	40.5	30.4	29.8	20.8	19.8
50 Hours	29.4	42.9	34.5	38.1	22.1	23.6
54 Hours	53.5	71.7	58.2	66.0	31.6	40.1
58 Hours	60.1	78.0	65.2	70.1	44.2	42.8
60 Hours	66.3	83.3	63.3	72.2	44.6	42.0
Averages	47.16	63.57	48.59	54.11	33.70	34.50

*Time after first insulin injection. A second injection was given 48 hours after the first.



BLOOD SUGAR RESPONSES AT 0.8 UNIT OF INSULIN PER KILOGRAM OF BODY WEIGHT

Figure 15

Changes in blood sugar presented here represent an average of two methods of sugar determination.

ways by research workers. In mice, sensitivity has been measured in terms of the amount of insulin required to cause convulsions[15]. Hereditary hypopituitary dwarf mice are much more sensitive to insulin by this measurement than are normal individuals. None of the cattle in this study went into convulsions; therefore, it was not possible to determine if dwarfs were more sensitive in this respect than normal appearing individuals. It has been shown that ruminants usually require huge doses of insulin administered over a long period of time before they will go into convulsions[6, 17].

Another means of measuring the sensitivity to insulin is to measure the degree of blood sugar fall in response to insulin injections. Rats with hypothalamic damage are much more sensitive to insulin when measured by this method than are those which are normal. This may be due to a decreased secretion of the growth hormone from the pituitary[19]. In the present study with cattle, as shown in Tables I and II and in Figures 15 and 16, dwarfs did show an increased blood sugar fall after insulin as compared to carriers and pedigree-clean animals. This was especially true when

the larger dosage of insulin was administered. Carrier animals were intermediate in this respect but differed only slightly from those which were pedigree-clean.

Insulin sensitivity may also be expressed as the area confined by the blood sugar curve and a line parallel to the abscissa which intersects the curve at the initial post-absorptive level[9]. Data presented in Figure 15 show very clearly that when the larger dosage of 0.8 unit of insulin was administered, the area of the blood sugar curve was larger for the dwarfs and smaller for the pedigree-clean animals, with the carriers intermediate. Thus, as measured in this way, the dwarfs and carriers were more sensitive to insulin than dwarf-free individuals. At the lower dosage of insulin, the dwarfs were still more sensitive to insulin as measured by this method, but there was little difference between carrier and pedigree-clean individuals. In fact, at the low dosage of insulin the carriers appeared less sensitive to insulin by this measurement than the pedigree-clean individuals.

Blood glucose areas were calculated for each of the individuals in the three genotypes and were analyzed statistically. The results of this analysis are given in Table IV and show a highly significant difference in glucose areas between genotypes, between periods of time after insulin injections and between the different dosages of insulin administered. Glucose areas averaged 1.21 times larger at the higher than the lower dosage of the hormone for all individuals combined. Of particular interest are the significant interactions involving genotype and dosage level and area changes after insulin injection in the same individuals. These further substantiate the conclusions based on the data in Figures 15 and 16 that animals of the three genotypes for the dwarf gene responded differently at the different levels of insulin administered.

The increased sensitivity of dwarfs to insulin indicates that the pituitary gland may not be functioning properly. An increase in insulin sensitivity in dogs has been noted after hypophysectomy[9]. Furthermore, there is evidence that the return of blood glucose to the initial level after insulin administration is dependent upon a release of glucose from the liver in response to the insulin induced hypoglycemia[4]. In turn, glycogen accumulation is known to be affected by hormones secreted by both the pituitary gland and the adrenal cortex[5].

Whether the apparent upset in carbohydrate metabolism in the "snorter" dwarf as found in this study is the direct or indirect function of the dwarf gene has not been determined. The fact that a normal amount of pituitary hormone seems to be present in the pituitary of the dwarf[13] in contrast to the indications that some of these hormones do not function normally suggests that something probably interferes with their release from the pituitary gland. Perhaps the hypothalamus may not be functioning normally in the dwarf.

TABLE III. Mean squares and significant differences between dosage levels, methods of blood sugar analysis, time intervals, genotypes, and the interactions involved

Source	DF	MS	F Ratio
Between dosage level (.8 and .3)	1	3811	19.73*
Between methods (Folin-Wu and Nelson-Somogyi)	1	11363	58.82*
Between time intervals (1 to 7)	6	26790	138.68*
Between genotypes (clean, carrier and dwarf)	2	38105	197.26*
Level × time	6	1718	8.89*
Level × method	1	200	1.03†
Time × method	6	120	1.62†
Level × genotype	2	2583	13.37*
Time × genotype	12	629	3.26*
Method × genotype	2	7162	37.07*
Level × time × method	6	72	.37†
Level × time × genotype	12	326	1.69†
Method × time × genotype	12	57	.29†
Level × method × genotype	2	2163	11.20*
Level × method × genotype × time	12	307	1.59†
Within	924	193	
Total	1007		

*Probability of chance occurrence less than .01.
†Differences not significant.

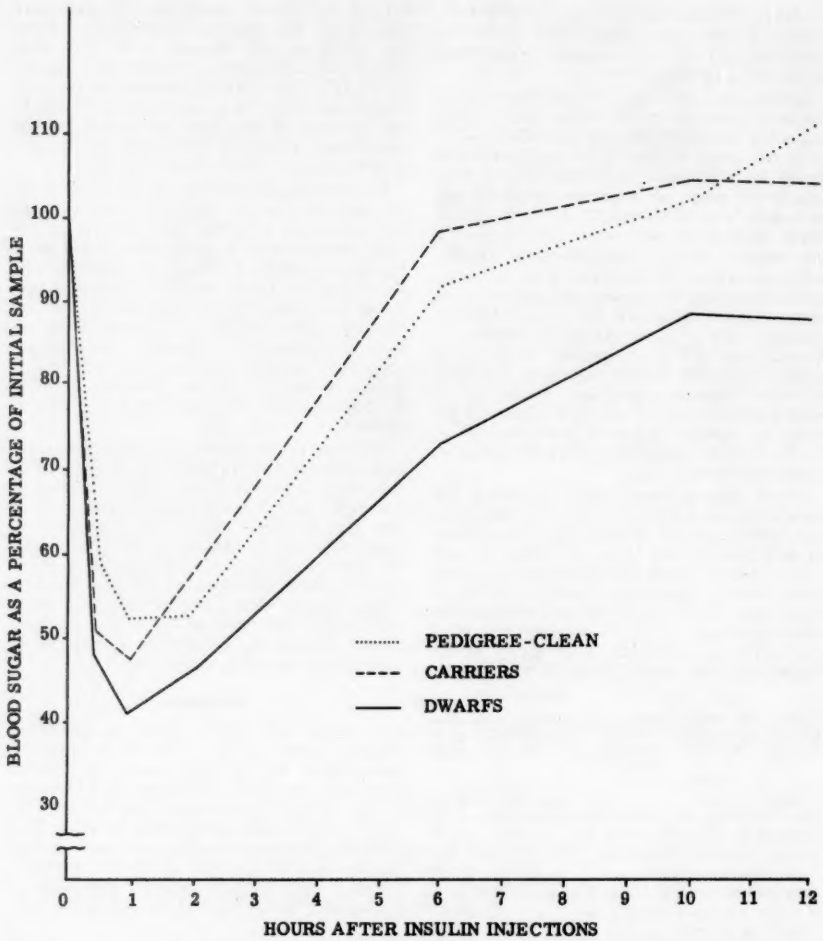
TABLE IV. Analysis of variance for blood sugar areas with data from both methods of blood sugar analysis combined

Source	DF	MS	F Ratio
Genotype	2	478	5.80†
Time	5	13622	165.17†
Level	1	4933	59.82†
Genotype × time	10	379	4.60†
Level × genotype	2	317	3.85*
Time × level	5	1562	18.94†
Level × time × genotype	10	169	2.05*
Within	828	82	
Total	863		

*Probability of chance occurrence less than .05.
†Probability of chance occurrence less than .01.

Summary

A study was made of the sensitivity of dwarfs, known carriers of the dwarf gene and homozygous normal beef cattle to insulin. Sensitivity to insulin was determined by measuring changes in the blood glucose level by two methods after insulin administration. Two different dosages of insulin were used—a high dosage level at the rate of 0.8 unit per kilogram of body weight followed within 48 hours by a lower dosage level of 0.3 unit in the same



BLOOD SUGAR RESPONSES AT 0.3 UNIT OF INSULIN PER KILOGRAM OF BODY WEIGHT

Figure 16

This injection was made 48 hours after the injection of 0.8 unit of insulin per kilogram of body weight. Changes in blood sugar represent an average of two methods of sugar determination.

animal. These studies indicated that the dwarfs were much more sensitive to insulin than were the normal animals or those which were known carriers of the dwarf gene. These results suggest that the gene responsible for "snorter" dwarfism is in some manner directly or indirectly concerned with carbohydrate metabolism and in the ability of the dwarf to cope with an insulin induced stress. Possibly the anterior pituitary and the adrenals are not functioning normally in the dwarf.

Blood sugar determinations as reported in this study were of little value in distinguishing between carriers of the dwarf gene and pedigree-clean animals because of the overlap between the two groups. The significant dif-

ferences between means for the different genotypes studied are of value, however, because they suggest a possible physiological effect of the dwarf gene.

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THE RELATION BETWEEN SEX AND SURVIVAL IN SWINE

D. F. Cox*

SWINE have an excess of males at birth, and there is a higher mortality in males during early life. Although this is similar to the results in man and many other species, it is not universal in all mammals. Studies by King[1] and, more recently, by Weir[3] have shown that the sex ratio at birth can be altered by selection. Furthermore, Weir[4] could not demonstrate any significant differential mortality of sexes through weaning. Knowledge of the factors responsible for such varying patterns of survival is presently incomplete.

Survival in swine, as in many other forms, is influenced adversely by inbreeding and, conversely, displays heterosis from crossbreeding. The mortality rates of males and females from purebred and from crossbred swine were investigated in data from the Iowa State University herd to determine whether the heterotic effects of crossbreeding were similar in each sex.

Data

The data came from a project at Iowa State University on evaluating seven breeds of swine commonly used in Iowa and the 42 possible crosses between these breeds. Purebred and crossbred individuals were raised concurrently at a single location in six separate seasons, spring or fall from 1954 to 1956. The purebred offspring were from crosses of purebred individuals from the same breed, and the crossbred offspring were from crosses of purebred individuals from different breeds. No crossbred individuals were used as parents. The number born dead in each litter and the age at death of each individual which died before reaching slaughter age were recorded. The individuals were grouped into nine groups according to age at death, beginning with those dead at birth and ending with those surviving at slaughter age. The data include 2,216 purebred and 2,353 crossbred births.

Results and Discussion

Life tables from birth to slaughter age for purebred and crossbred males and females are given in Table I. For comparison, these tables were constructed on the basis of 10,000 births. The actual numbers in the four groups ranged from 1,089 to 1,239. These tables cover the life of the individual up to sexual maturity. The pigs were weaned at approximately 56 days. In most cases, the males were castrated at weaning, but some were castrated at 21 days. A few purebred males were saved as breeding stock, while all crossbred males were castrated. This fact could influence comparisons of the mortality rates after 21 days, but before that time the conditions were comparable in the four groups.

The crossbred males showed a slightly higher mortality than the purebred males at birth and during the first three days of life. From three to 70 days of age, the crossbred males had lower mortality than the purebred males. The course of mortality in the females is somewhat similar, except that differences between the purebred and crossbred mortality rates after birth are

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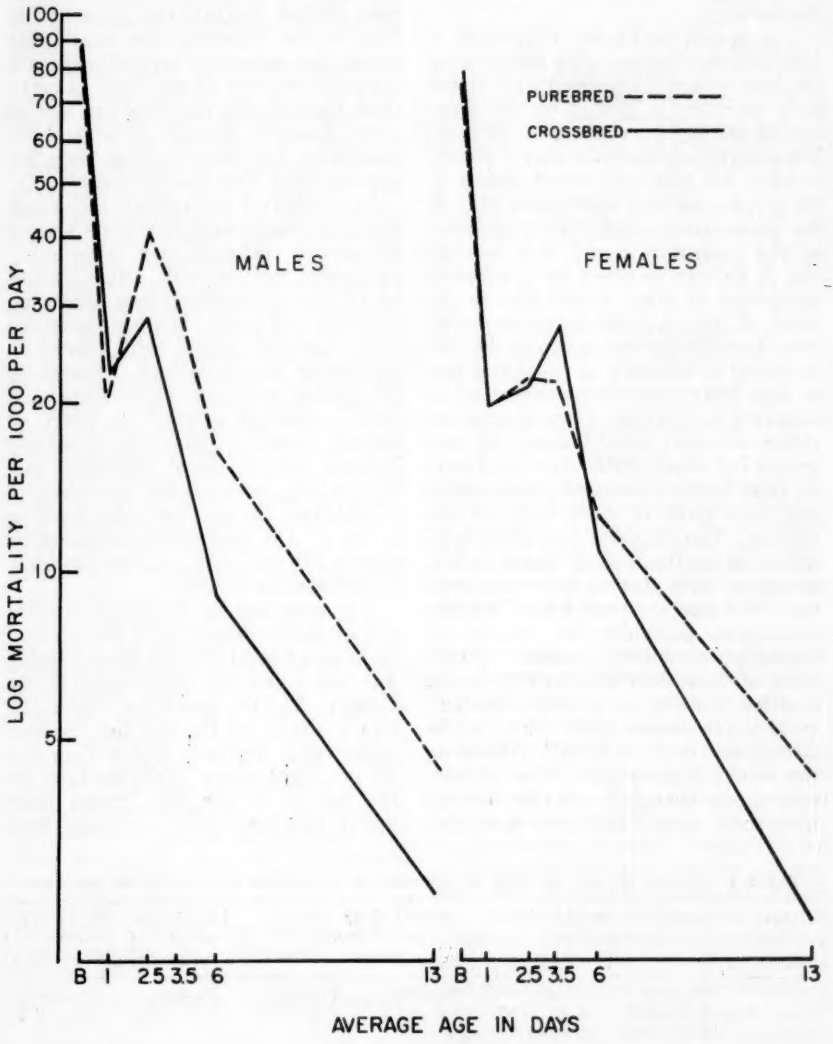


MORTALITY RATES

Figure 17

The logarithm of the daily mortality is plotted for the averages of the age intervals up to 20 days. The parallelism among the four lines is apparent. For the intervals after two days, the difference between the mortality rates of purebred and crossbred males is distinctly larger than the difference of purebred and crossbred females.

much smaller than those found between the males.

The graphs in Figure 17 provide a comparison of the mortality rates among the four groups. The logarithm of the daily mortality is plotted for the averages of the age intervals up to 20 days. The last two age intervals were excluded to allow for more convenient scaling in the graphs, for they contributed little to the comparisons made. The logarithm of daily mortality would be a straight line if the rate declined by a constant percentage of time. Parallelism in the plots of the logarithms of mortality rates from two groups indicates that the mortality is changing at the same rate in both groups, but the average level of mortality is different. These graphs are rather strikingly parallel among the four groups but depart distinctly from linearity from birth to two days for the males and from birth to three days for the females. This indicates a reduced mortality during the first 48 hours of life, compared with that at birth and with that after two or three days. The circumstances governing the records on mortality in swine probably permit some of those individuals dying in the first few minutes or, in some instances, even a few hours after birth to be classed as "dead at birth." However, the results indicate that those individuals strong enough to survive through birth and a short period immediately following are subject to lower mortality rates during the first two days of life than in the following few days. The young pigs apparently are endowed with sufficient reserves of nutrients to carry them through this period in the face of even strongly adverse environmental conditions, but these reserves soon are depleted, and then mortality rises.

In crossbred individuals the slight excess of mortality at birth and shortly afterwards, although not statistically significant, may be real. Other studies on this same population have shown no essential difference between purebred and crossbred litters with regard to number or weight at birth. Therefore, the higher mortality in crossbreds at birth cannot be charged to larger or heavier litters. Some incompatibility between the crossbred individual and the uterine or postnatal environment provided by the purebred dam could be a factor. The possibilities of hemolytic disease through such an incompatibility is discussed by Roberts[2].

The most striking feature of the data is that the improvement in the survival of males achieved through crossbreeding was larger than that experienced by the females. On the graph in Figure 17, this is shown by the line for purebred males being distinctly higher than that for crossbred males, while the lines for the females in the two groups more nearly coincide. This is opposite from

TABLE I. Life table for each sex within the purebred and the crossbred group (of 10,000 pigs born)

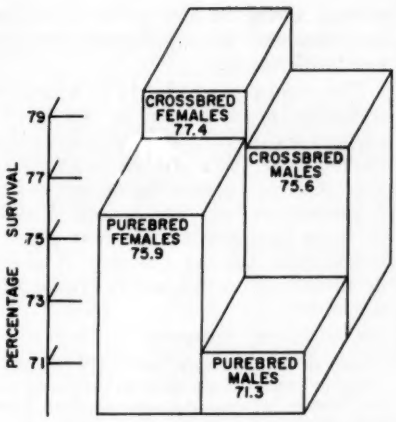
Age interval Days	Number alive at beginning of age interval l_x				Number dying during the age interval d_x				Mortality per 1000 per day $q_x = \frac{(d_x)}{(l_x)} \left(\frac{1000}{\text{(days in interval)}}\right)$			
	Purebred		Crossbred		Purebred		Crossbred		Purebred		Crossbred	
	♂♂ (1134)*	♀♀ (1082)	♂♂ (1239)	♀♀ (1114)	♂♂	♀♀	♂♂	♀♀	♂♂	♀♀	♂♂	♀♀
Birth	10000	10000	10000	10000	802	730	859	772	80.2	73.0	85.9	77.2
0-2	9198	9270	9141	9228	379	361	405	359	20.6	19.4	22.2	19.4
2-3	8819	8909	8736	8869	362	194	252	188	41.0	21.8	28.8	21.2
3-4	8457	8715	8484	8681	255	185	146	234	30.2	21.2	17.2	27.0
4-6	8202	8530	8338	8447	264	212	146	179	16.1	12.4	8.8	10.6
6-20	7938	8318	8192	8268	415	352	299	269	4.4	4.2	2.6	2.3
20-70	7523	7966	7893	7999	327	305	219	171	0.9	0.8	0.6	0.4
70-150	7196	7661	7674	7828	62	74	114	90	0.1	0.1	0.2	0.1
Survivors	7134	7587	7560	7738								

*Actual numbers born.

what might be expected if the sex chromosomes were important in causing heterosis.

Whether these differences are other than chance ones can be tested by comparing the number of individuals dying in each age interval to 150 days, including the number surviving at 150 days among the various groups. The nine age intervals and the four classifications of individuals form a two-way contingency table with 24 degrees of freedom to test the independence of rows and columns. The total chi-square from this table is partitioned in Table II. The results indicate that the distribution of deaths between purebred and crossbred males differs more than is likely to have occurred by chance. On the other hand, while the distribution of deaths between the sexes in general and between the females in the crossbred and purebred groups may have been really different, the present data are insufficient to demonstrate the statistical significance of this.

The percentages of survival to slaughter age in the four groups of individuals can be compared graphically by the three-dimensional model in Figure 18. The columns rising from the horizontal plane represent the percentage of those reaching slaughter age in the various classes. The lack of additivity in the effect of sex and of type of breeding on survival is evident in the diagram. The front face of the diagram, representing the sexes of the purebred group, shows a difference of over twice that seen in the opposite face of the diagram which represents the sex differences among crossbreds. The sides of the model contrast the difference in heterosis between the two sexes. Crossbreeding increases the percentage surviving in both sexes, but the increase it causes in males is almost three times that in females.



SURVIVAL TO SLAUGHTER AGE

Figure 18

The model illustrates the lack of additivity between the effects of sex and type of breeding on survival to slaughter age. The sex difference is over twice as large in the purebred as in the crossbred groups. The influence of crossbreeding is almost three times as large in males as in females.

Besides the males responding more than females to crossbreeding, the rates of mortality varied more from season to season in the males than in the females. Heterogeneity of total mortality from birth to slaughter age over the six seasons was tested by chi-squares within the four classes. These chi-squares based on five degrees of freedom are shown in Table III. The figures show that the heterogeneity in male mortality is unlikely to be a mere chance occurrence. Apparently, the males, especially the purebred males, are not able to adapt

TABLE II. Chi-square values for comparing the distribution of deaths in four types of individuals over nine age intervals

Comparison	d.f.	Chi-square	Probability
♂♂ vs. ♀♀	8	10.38	.3 to .2
Crossbred ♂♂ vs. purebred ♂♂	8	18.77	.02 to .01
Crossbred ♀♀ vs. purebred ♀♀	8	6.77	.7 to .5

TABLE III. Chi-squares (5 d.f.) for the heterogeneity of death rates among six seasons in four types of individuals

	Males		Females	
	Purebred	Crossbred	Purebred	Crossbred
Chi-square	15.16	9.92	4.45	5.08
Probability	<.01	.05 to .02	.5 to .3	.5 to .3

as well as the females to the changing environmental factors included in seasonal differences.

The results point clearly to a lack of additivity in the effects of crossbreeding and sex on early mortality in swine. Furthermore, the variability in mortality over seasons appears to be somewhat dependent on sex. The biological basis of these phenomena and their genetic implications are not obvious. A more detailed study of the factors involved is warranted.

Summary

Data on 2,216 purebred and 2,353 crossbred births in swine were studied to determine the relationship between sex and survival from birth to slaughter age in these two breeding groups. The heterosis, expressed as reduced mortality, was almost three times as large in the males as in the females. The difference between male and female mortality was over twice as large in the purebreds as it was in the crossbreds. The variability in mortality over seasons was more extreme among the males, especially in the purebred population.

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